METHODS AND COMPOSITIONS FOR DETECTING SARS VIRUS AND OTHER INFECTIOUS AGENTS

BACKGROUND OF THE INVENTION

Since November of 2002, a disease called severe acute respiratory syndrome (SARS) has been reported in twenty two countries around the world. WHO has reported 6,054 cumulative cases of SARS and 417 death among infected people as of May 2, 2003. For the same period, China has reported 3,788 cumulative cases of SARS and 181 deaths among infected people.

The main symptoms for SARS patients include fever (greater than 38°C), headache, body aches. After 2-7 days of illness, patients may develop a dry, nonproductive cough that may be accompanied with breathing difficulty.

Based on findings from Hong Kong, Canada, and U.S., a previously unrecognized coronaviruse has been identified as the cause of SARS. Researchers have found that SARS coronaviruse is a positive chain RNA virus which replicates without DNA intermediate step and uses standard codon (Marra et al., Science 2003 May 1; (epub ahead of print); and Rota et al., Science 2003 May 1, (epub ahead of print)).

SARS coronaviruse is a newly discovered virus which has not been previously detected in human or animals. The genome structure of SARS coronaviruse is very similar to other coronaviruse. The genome of SARS coronaviruse is 30 K base pairs in length and the genome is considered very large for a virus. The genome of SARS coronaviruse encodes RNA polymerase (polymerase 1a and 1b), S protein (spike protein), M protein (membrane protein), and N protein (nucleocapsid protein), etc.

Currently, there are three types of detection methods for SARS coronaviruse: immunological methods (e.g., ELISA), reverse transcriptase polymerase chain reaction (RT-PCR) tests, and cell culture methods.

There are significant drawbacks of the above three detection methods. For example, ELISA can reliably detect antibodies from serum of SARS patients. However, those antibodies can only be detected twenty one days after development of symptoms. Cell culture methods have a relative long detection cycle and can be applied only to

limited conditions. In addition, cell culture methods can only detect existence of alive virus.

The key step of preventing the spread of SARS coronaviruse is early diagnosis and early quarantine and treatment. RT-PCR is the only existing method that allows detection of nucleic acid of SARS coronaviruse. However, RT-PCR cannot eliminate infected patient before SARS virus expression, and detection rate for RT-PCR is low. The detection process requires expensive real time PCR equipment. Thus, RT-PCR cannot satisfy the need of early clinical screening and diagnosis. There exists a need in the art for a quick, sensitive and accurate diagnosis of the severe acute respiratory syndrome (SARS). The present invention address this and other related needs in the art.

BRIEF SUMMARY OF THE INVENTION

The current method for clinical diagnosis is mainly based on symptoms such as fever, shadows on patient's lung, dry cough, and weakness in patient's arms and legs. However, these symptoms are not specific for SARS; other pathogens can cause the same or similar symptoms. For example, regular pneumonia caused by *Chlamydia pneumoniae* and *Mycoplasma pneumoniae* also generates shadows on patient's lung; fever and cough are also associated with influenza; and similar symptoms are also associated with infection of the upper respiratory tract caused by human coronaviruse 229E and OC43. Thus, diagnosis for SARS solely based on the symptoms of the patient is problematic.

Current clinical data indicate that many suspected SARS cases actually did not have infection by SARS virus, and instead, had infection by other pathogens. Thus, there is a need to develop a method for simultaneous detection of SARS and other pathogens that cause symptoms similarly to SARS. Such method would provide quick screening of suspected cases in order to reduce probability of diagnostic errors, to allow timely and adequate treatment, and to avoid unnecessary panic and medical waste. Patients infected with SARS virus are more susceptible to other pathogens due to decreased immunity caused by SARS virus. It is possible that SARS patients are also infected with other pathogens that generate symptoms similar to SARS. For example, if

a patient is infected with both SARS and *Mycoplasma pneumoniae*, treatment with medicine only for SARS will not make symptoms disappear immediately. In this situation, a simultaneous detection of infection by both pathogens would allow immediate and effective treatment of patients for both pathogens. A biochip-based diagnosis is a fast and low cost method for high throughput simultaneous screening of multiple samples. Thus, one objective of the invention is to provide a biochips for simultaneous detection of SARS virus and other pathogens that cause SARS-like symptoms.

Clinical data also indicate that those SARS patients infected with other pathogens (pathogens that severely interfere and obstruct immunity, such as hepatitis B and HIV) have aggravated symptoms and high probability of infecting others (these patients are called "super-spreaders"). Proper detection of such patients would allow adequate treatment and timely quarantine of patients. Thus, another objective of the invention is to provide a nucleic acid microarray for simultaneous detection of SARS virus and other pathogens that aggravates symptoms of SARS.

In one aspect, the present invention is directed to a chip for assaying for a coronaviruse causing the severe acute respiratory syndrome (SARS-CoV) and a non-SARS-CoV infectious organism, which chip comprises a support suitable for use in nucleic acid hybridization having immobilized thereon an oligonucleotide probe complementary to a nucleotide sequence of SARS-CoV genome, said nucleotide sequence comprising at least 10 nucleotides, and one or more of the following oligonucleotide probe(s): a) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism causing SARS-like symptoms, said nucleotide sequence comprising at least 10 nucleotides; b) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism damaging an infectious host's immune system, said nucleotide sequence comprising at least 10 nucleotides; or c) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV coronaviridae virus, said nucleotide sequence comprising at least 10 nucleotides.

In some embodiments, the chip of the invention comprises a support suitable for use in nucleic acid hybridization having immobilized thereon at least two oligonucleotide probes complementary to at least two different nucleotide sequences of SARS-CoV genome, each of said two different nucleotide sequences comprising at least 10 nucleotides.

In some embodiments, the non-SARS-CoV infectious organism causing SARS-like symptoms is selected from the group consisting of a human coronaviruse 229E, a human coronaviruse OC43, a human enteric coronaviruse, an influenza virus, a parainfluenza virus, a respiratory sncytical virus, a human metapneumovirus, a rhinovirus, an adenoviruse, a mycoplasma pneumoniae, a chlamydia pneumoniae, a measles virus and a rubella virus.

In some embodiments, the non-SARS-CoV infectious organism damaging an infectious host's immune system is selected from the group consisting of a hepatitis virus, a transfusion transmitting virus (TTV), a human immunodeficiency virus (HIV), a parvovirus, a human cytomegalovirus (HCMV), an Epstein-Barr virus (EBV) and a tre-ponema palidum.

In another aspect, the present invention is directed to a method for assaying for a SARS-CoV and a non-SARS-CoV infectious organism in a sample, which methods comprises: a) providing an above-described chip; b) contacting said chip with a sample containing or suspected of containing a nucleotide sequence of a SARS-CoV and a non-SARS-CoV infectious organism under conditions suitable for nucleic acid hybridization; and c) assessing hybrids formed between said nucleotide sequence of said SARS-CoV or said non-SARS-CoV infectious organism, if present in said sample, and said oligonucleotide probe complementary to a nucleotide sequence of said SARS-CoV genome or said oligonucleotide probe complementary to a nucleotide sequence of said non-SARS-CoV infectious organism genome, whereby detection of one or both of said hybrids indicates the presence of said SARS-CoV and/or said non-SARS-CoV infectious organism in said sample.

In some embodiments, the SARS-CoV is assayed by: a) providing a chip comprising a support suitable for use in nucleic acid hybridization having immobilized

thereon at least two oligonucleotide probes complementary to at least two different nucleotide sequences of SARS-CoV genome, each of said two different nucleotide sequences comprising at least 10 nucleotide; b) contacting said chip with a sample containing or suspected of containing a SARS-CoV nucleotide sequence under conditions suitable for nucleic acid hybridization; and c) assessing hybrids formed between said SARS-CoV nucleotide sequence, if present in said sample, and said at least two oligonucleotide probes complementary to two different nucleotide sequences of SARS-CoV genome, respectively, to determine the presence, absence or amount of said SARS-CoV in said sample, whereby detection of one or both said hybrids indicates the presence of said SARS-CoV in said sample.

By using multiple hybridization probes, the present methods reduce the occurrence of false negative results compared to a test based on a single hybridization probe as the chance of simultaneous mutations of the multiple hybridization targets is much smaller than the chance of a mutation in the single hybridization target. When other preferred embodiments are used, *e.g.*, a negative control probe and a blank spot on the chip, the chance of a false positive result can also be reduced. The inclusion of more preferred embodiments, *e.g.*, an immobilization control probe and a positive control probe, on the chip can provide further validation of the assay results. The use of preferred sample preparation procedures, RNA extraction procedures and amplification procedures can further enhance the sensitivity of the present methods.

In still another aspect, the present invention is directed to an oligonucleotide primer for amplifying a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43, which oligonucleotide primer comprises a nucleotide sequence that: a) hybridizes, under high stringency, with a target nucleotide sequence of influenza A virus, influenza B virus, human metapneumovirus, human adenovirus, human coronaviruse 229E or human coronaviruse OC43, or a complementary strand thereof, that is set forth in Tables 1-6; or b) has at least 90% identity to a target nucleotide sequence of influenza A virus, influenza B virus, human metapneumovirus, human

adenovirus, human coronaviruse 229E or human coronaviruse OC43 comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Tables 1-6.

Table 1. Exemplary Influenza A Virus Primers

Id	Sequence
PMIA_00001	TTTGTGCGACAATGCTTCA
PMIA_00002	GACATTTGAGAAAGCTTGCC
PMIA_00003	AGGGACAACCTNGAACCTGG
PMIA_00004	AGGAGTTGAACCAAGACGCATT
PMIA_00005	ACCACATTCCCTTATACTGGAG
PMIA_00006	TTAGTCATCATCTTTCTCACAACA
PMIA_00007	ACAAATTGCTTCAAATGAGAAC
PMIA_00008	TGTCTCCGAAGAAATAAGATCC
PMIA_00009	GCGCAGAGACTTGAAGATGT
PMIA_00010	CCTTCCGTAGAAGGCCCT

Table 2. Exemplary Influenza B Virus Primers

Id	Sequence
PMIB_00001	CACAATGGCAGAATTTAGTGA
PMIB_00002	GTCAGTTTGATCCCGTAGTG
PMIB_00003	CAGATCCCAGAGTGGACTCA
PMIB_00004	TGTATTACCCAAGGGTTGTTAC
PMIB_00005	GATCAGCATGACAGTAACAGGA
PMIB_00006	ATGTTCGGTAAAAGTCGTTTAT
PMIB_00007	CCACAGGGGAGATTCCAAAG
PMIB_00008	GACATTCTTCCTGATTCATAATC
PMIB_00009	CAAACAACGGTAGACCAATATA
PMIB_00010	AGGTTCAGTATCTATCACAGTCTT
PMIB_00011	ATGTCCAACATGGATATTGAC

Id	Sequence
PMIB_00012	GCTCTTCCTATAAATCGAATG
PMIB_00013	TGATCAAGTGATCGGAAGTAG
PMIB_00014	GATGGTCTGCTTAATTGGAA
PMIB_00015	ACAGAAGATGGAGAAGGCAA
PMIB_00016	ATTGTTTCTTTGGCCTGGAT

Table 3. Exemplary Human Metapneumovirus Primers

Id	Sequence
PMM_00001	CATCCCAAAAATTGCCAGAT
PMM_00002	TTTGGGCTTTGCCTTAAATG
PMM_00003	ACACCCTCATCATTGCAACA
PMM_00004	GCCCTTCTGACTGTGGTCTC
PMM_00005	CGACACAGCAGCAGGAATTA
PMM_00006	TCAAAGCTGCTTGACACTGG
PMM_00007	CAAGTGCGACATTGATGACC
PMM_00008	TAATTCCTGCTGCTGTGTCG
PMM_00009	GCGACTGTAGCACTTGACGA
PMM_000010	TCATGATCAGTCCCGCATAA
PMM_000011	TGTTTCAGGCCAATACACCA
PMM_000012	TCATGATCAGTCCCGCATAA
PMM_000013	TCATGGGTAATGAAGCAGCA
PMM_000014	GGAGTTTTCCCATCACTGGA
PMM_000015	TCCAGTGATGGGAAAACTCC
PMM_000016	TGTTGAGCTCCTTTGCCTTT

Table 4. Exemplary Human Adenovirus Primers

Id	Sequence
PMAd1_00001	TGGCGGTATAGGGGTAACTG

Id	Sequence
PMAd1_00002	ATTGCGGTGATGGTTAAAGG
PMAd1_00003	TTTTGCCGATCCCACTTATC
PMAd1_00004	GCAAGTCTACCACGGCATTT
PMAd2_00001	CTCCGTTATCGCTCCATGTT
PMAd2_00002	AAGGACTGGTCGTTGGTGTC
PMAd2_00003	AAATGCCGTGGTAGATTTGC
PMAd2_00004	GTTGAAGGGGTTGACGTTGT
PMAd3_00001	TCCTCTGGATGGCATAGGAC
PMAd3_00002	TGTTGGTGTTAGTGGGCAAA
PMAd3_00003	ACATGGTCCTGCAAAGTTCC
PMAd3_00004	GCATTGTGCCACGTTGTATC
PMAd4_00001	CGCTTCGGAGTACCTCAGTC
PMAd4_00002	CTGCATCATTGGTGTCAACC
PMAd4_00003	GGCACCTTTTACCTCAACCA
PMAd4_00004	TCTGGACCAAGAACCAGTCC
PMAd5_00001	GGCCTACCCTGCTAACTTCC
PMAd5_00002	ATAAAGAAGGGTGGGCTCGT
PMAd5_00003	ATCGCAGTTGAATGCTGTTG
PMAd5_00004	GTTGAAGGGGTTGACGTTGT
PMAd7_00001	ACATGGTCCTGCAAAGTTCC
PMAd7_00002	GATCGAACCCTGATCCAAGA
PMAd7_00003	AACACCAACCGAAGGAGATG
PMAd7_00004	CCTATGCCATCCAGAGGAAA
PMAd11_00001	CAGATGCTCGCCAACTACAA
PMAd11_00002	AGCCATGTAACCCACAAAGC
PMAd11_00003	ACGGACGTTATGTGCCTTTC
PMAd11_00004	GGGAATATTGGTTGCATTGG
PMAd21_00001	ACTGGTTCCTGGTCCAGATG
PMAd21_00002	AGCCATGTAACCCACAAAGC

Id	Sequence
PMAd21_00003	CTGGATATGGCCAGCACTTT
PMAd21_00004	CACCTGAGGTTCTGGTTGGT
PMAd23_00001	TAATGAAAAGGGCGGACAAG
PMAd23_00002	GCCAATGTAGTTTGGCCTGT
PMAd23_00003	AACTCCGCGGTAGACAGCTA
PMAd23_00004	CGTAGGTGTTGGTGTTGGTG

Table 5. Exemplary HCoV-OC229E Primers

Id	Sequence
PMV_a0053	TCACTTGCTTCCGTTGAGGTTGGGCTGGCGGTTTAGAGTTGA
PMV_a0054	GGTTTCGGATGTTACAGCGTGTGCGACCGCCCTTGTTTATGG
PMV_a0055	TCACTTGCTTCCGTTGAGGGCGTTGTTGGCCTTTTTCTTGTCT
PMV_a0056	GGTTTCGGATGTTACAGCGTGCCCGGCATTATTTCATTGTTCTG
PMV_a0057	TCACTTGCTTCCGTTGAGGACAAAAGCCGCTGGTGGTAAAG
PMV_a0058	GGTTTCGGATGTTACAGCGTCAGAAATCATAACGGGCAAACTCA
PMV_a0059	TCACTTGCTTCCGTTGAGGAAGAGTTATTGCTGGCGTTGTTGG
PMV_a0060	GGTTTCGGATGTTACAGCGTGCCCGGCATTATTTCATTGTTCTG
PMV_b0053	TTGGGCTGGCGGTTTAGAGTTGA
PMV_b0054	GTGCGACCGCCCTTGTTTATGG
PMV_b0055	GCGTTGTTGGCCTTTTTCTTGTCT
PMV_b0056	GCCCGGCATTATTTCATTGTTCTG
PMV_b0057	ACAAAAGCCGCTGGTGGTAAAG
PMV_b0058	CAGAAATCATAACGGGCAAACTCA
PMV_b0059	AAGAGTTATTGCTGGCGTTGTTGG
PMV_b0060	GCCCGGCATTATTTCATTGTTCTG

Table 6. Exemplary HCoV-OC43 Primers

Id	Sequence

Id	Sequence
PMV_a0061	TCACTTGCTTCCGTTGAGGTTGGGGTGATGGGTTTCAGATTAA
PMV_a0062	GGTTTCGGATGTTACAGCGTCTCGGGAAGATCGCCTTCTTCTA
PMV_b0061	TTGGGGTGATGGGTTTCAGATTAA
PMV_b0062	CTCGGGAAGATCGCCTTCTTCTA

In yet another aspect, the present invention is directed to a kit for amplifying a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43, which kit comprises: a) a primer described above; and b) a nucleic acid polymerase that can amplify a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43 using said primer.

In yet another aspect, the present invention is directed to an oligonucleotide probe for hybridizing to a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43, which oligonucleotide probe comprises a nucleotide sequence that: a) hybridizes, under high stringency, with a target nucleotide sequence of influenza A virus, influenza B virus, human metapneumovirus, human adenovirus, human coronaviruse 229E or human coronaviruse OC43, or a complementary strand thereof, that is set forth in Tables 7-12; or b) has at least 90% identity to a target nucleotide sequence of influenza A virus, influenza B virus, human metapneumovirus, human adenovirus, human coronaviruse 229E or human coronaviruse OC43, or a complementary strand thereof, that is set forth in Tables 7-12.

Table 7. Exemplary Influenza A Virus Probes

Id	Sequence
PBIA_00001	TTTAGAGCCTATGTGGATGGATTCRAACCGAACGGCTGCATTGAGGGCAAGCTTTCTCAAATGTC
PBIA_00002	ACAATTGAAGAAAGATTTGAAATCACTGGAACCATGCGCAGGCTTGCCGACCAAAGTCTCCCACCGAACT

Id	Sequence
PBIA_00003	AGCAATNGAGGAGTGCCTGATTAANGATCCCTGGGTTTTGCTNAATGC
PBIA_00004	CCATACAGCCATGGAACAGGAACAGGATACACCATGGACACAGTCAACAGAACACANCAATATTCAGAAA
PBIA_00005	GGGCGGGGAGTCTTCGAGCTCTCNGACGAAAAGGCAACGAACCCGATCGTGCC
PBIA_00006	GATCTNGAGGCTCTCATGGAATGGCTAAAGACAAGACCAATCCTGTCACCTCTGACTAA

Table 8. Exemplary Influenza B Virus Probes

Id	Sequence
PBIB_00001	GCTGGGAAATAGCATGGAACTGATGATATTCAGCTACAATCAAGACTATTCGTTAAGTAATGAATCCTCA
PBIB_00002	TCTGTTCCAGCTGGTTTCTCCAATTTTGAAGGAATGAGGAGCTACATAGACAATATAGATCCTAAAGGAG
PBIB_00003	TTACAACCATGAGCTACCAGAAGTTCCATATAATGCCTTTCTTCTAATGTCTGATGAATTGGGGCTGGCC
PBIB_00004	ACAAATAAGATCCAAATGAAATGGGGAATGGAAGCTAGAAGATGTCTGCTTCAATCAATGCAACAAATGG
PBIB_00005	GAGGGAATGTATTCTGGAATAGANGAATGTATTAGTAACAACCCTTGGGTAATACAGAGTGCATACTGGT
PBIB_00006	CTACCGTGTTGGGAGTAGCCGCACTAGGTATCAAAAACATTGGAAACAAAGAATACTTATGGGATGGACT
PBIB_00007	GGCTATGACTGAAAGAATAACCAGAGACAGCCCAATTTGGTTCCGGGATTTTTGTAGTATAGCACCGGTC
PBIB_00008	ACTGATCAGAGGAACATGATTCTTGAGGAACAATGCTACGCTAAGTGTTGCAACCTTTTTGAGGCCTGTT
PBIB_00009	AAAATCCCTTTGTNGGACATTTGTCTATTGAGGGCATCAAAGANGCAGATATAACCCCAGCACATGGTCC
PBIB_00010	CTTGGAATACAAGGGAATACAACTTAAAACAAATGCTGAAGACATAGGAACCAAAGGCCAAATGTGCTCA
PBIB_00011	GTGGCAGGAGCAACATCAGCTGAGTTCATAGAAATGCTACACTGCTTACAAGGTGAAAATTGGAGACAAA
PBIB_00012	GGAACCCATCCCCGGAAAGAGCAACCACAAGCAGTGAAGCTGATGTCGGAAGGAAAACCCAAAAGAAACA
PBIB_00013	CTGTTTCCAAAGATCAAAGGCACTAAAAAGAGTTGGACTTGACCCTTCATTAATCAGTACCTTTGCAGGA
PBIB_00014	AGAGTTTTGTCTGCATTAACAGGCACAGAATTCAAGCCTAGATCAGCATTAAAATGCAAGGGTTTCCATG
PBIB_00015	GAGGGACGTGATGCAGATGTCAAAGGAAATCTACTCAAGATGATGAATGACTCAATGGCTAAGAAAACCA
PBIB_00016	CCTATCAGGAATGGGAACAACAGCAACAAAAAAGAAAGGCCTGATTCTAGCTGAGAGAAAAATGAGAAGA
PBIB_00017	GCAAGTCAAAAGAATGGGGAAGGAATTGCAAAGGATGTAATGGAAGTGCTAAAGCAGAGCTCTATGGGAA

Table 9. Exemplary Human Metapneumovirus Probes

Id	Sequence
PBM_00001	AAAAGTGTATCACAGAAGTTTGTTCATTGAGTATGGCAAAGCATTAGGCTCATCATCTACAGGCAGCAAA

Id	Sequence
PBM_00002	GAAAGTCTATTTGTTAATATATTCATGCAAGCTTATGGAGCCGGTCAAACAATGCTAAGGTGGGGGGTCA
PBM_00003	ACGCTGTTGTGTGGAGAAATTCTGTATGCTAAACATGCTGATTACAAATATGCTGCAGAAATAGGAATAC
PBM_00004	TTAAGGAATCATCAGGTAATATCCCACAAAATCAGAGGCCCTCAGCACCAGACACACCCATAATCTTATT
PBM_00005	TGAGCAATCAAAGGAGTGCAACATCAACATATCCACTACAAATTACCCATGCAAAGTCAGCACAGGAAGA
PBM_00006	CTGTTCCATTGGCAGCAACAGAGTAGGGATCATCAAGCAGCTGAACAAAGGTTGCTCCTATATAACCAAC
PBM_00007	ACTTAATGACAGATGCTGAACTAGCCAGGGCCGTTTCTAACATGCCGACATCTGCAGGACAAATAAAATT
PBM_00008	AAAAAAAGGGAAACTATGCTTGCCTCTTAAGAGAAGACCAAGGGTGGTATTGTCAGAATGCAGGGTCAAC
PBM_00009	GAAAAGAACACACCAGTTACAATACCAGCATTTATCAAATCGGTTTCTATCAAAGAGAGTGAATCAGCCA
PBM_00010	CAAATCAGTTGGCAAAAAAACACATGATCTGATCGCATTATGTGATTTTATGGATCTAGAAAAGAACACA
PBM_00011	CAGCTAAAGACACTGACTATAACTACTCTGTATGCTGCATCACAAAGTGGTCCAATACTAAAAGTGAATG
PBM_00012	AAAAGAACACACCAGTTACAATACCAGCATTTATCAAATCGGTTTCTATCAAAGAGAGTGAATCAGCCAC
PBM_00013	CTATTATAGGAGAAAAAGTGAACACTGTATCTGAAACATTGGAATTACCTACTATCAGTAGACCCACCAA
PBM_00014	AAGTTAGCATGGACAGACAAAGGTGGGGCAATCAAAACTGAAGCAAAGCAAACAATCAAAGTTATGGATC
PBM_00015	CAGGAAAATACACAAAGTTGGAGAAAGATGCTCTAGACTTGCTTTCAGACAATGAAGAAGAAGATGCAGA
PBM_00016	CTAATAGCAGACATAATAAAAGAAGCCAAGGGAAAAGCAGCAGAAATGATGGAAGAAGAAATGAACCAGC

Table 10. Exemplary Human Adenovirus Probes

Id	Sequence
PBAd_00001	CTGACACCTACCAAGGTATAAAATCAAACGGAAACGGTAATCCTCAAAACTGGACCAAAAATGACGATTT
PBAd_00002	TCCTCTACTCCAACATTGCACTGTACCTGCCTGACAAGCTAAAATACACTCCTACAAATGTGGAAATATC
PBAd_00003	GCTATCGGAGGCAGAGTACTAAAAAAGACTACTCCCATGAAACCATGCTACGGATCGTATGCCAGACCTA
PBAd_00004	AGTATTGTTTTGTACAGTGAGGATGTTAATATGGAAACTCCTGATACTCACATTTCATACAAACCAAGCA
PBAd_00005	GGGAAACGATCTTAGAGTTGACGGGGCTAGCATTAAGTTTGACAGCATTTGTCTTTACGCCACCTTCTTC
PBAd_00006	TTGCCATTAAAAACCTCCTCCTCCTGCCAGGCTCATATACATATGAATGGAACTTCAGGAAGGATGTTAA
PBAd_00007	TTGCAACACGTAATGAAATAGGAGTGGGTAACAACTTTGCCATGGAAATTAACCTAAATGCCAACCTATG
PBAd_00008	TTGGGGTAACTGACACCTATCAAGCTATTAAGGCTAATGGCAATGGCTCAGGCGATAATGGAGATATTAC
PBAd_00009	AGGTATCAAGGCATTAAAGTTAAAACCGATGACGCTAATGGATGGGAAAAAGATGCTAATGTTGATACAG
PBAd_00010	GAGAAGTTTTCTGTACTCCAATGTGGCTTTGTACCTTCCAGATGTTTACAAGTACACGCCACCTAACATT

Id	Sequence
PBAd_00011	ATCAGTCATTTAACGACTACCTCTCTGCAGCTAACATGCTTTACCCCATTCCTGCCAATGCAACCAACAT
PBAd_00012	CTACTTCGTATATTCTGGATCTATTCCCTACCTGGATGGCACCTTTTACCTTAACCACACTTTCAAGAAG
PBAd_00013	ACCTGCCAGTGGAAGGATGCTAACAGCAAAATGCATACCTTTGGGGTAGCTGCCATGCCAGGTGTTACTG
PBAd_00014	ATAGAAGCTGATGGGCTGCCTATTAGAATAGATTCAACTTCTGGAACTGACACAGTAATTTATGCTGATA
PBAd_00015	TTGAAATTAAGCGCACCGTGGACGGCGAGGGGTACAACGTGGCCCAGTGCAACATGACCAAGGACTGGTT
PBAd_00016	CGGCAACGACCGGCTCCTGACGCCCAACGAGTTTGAAATTAAGCGCACCGTGGACGGCGAGGGGTACAAC
PBAd_00017	CTCCAGTAACTTTATGTCCATGGGCGCACTCACAGACCTGGGCCAAAACCTTCTCTACGCCAACTCCGCC
PBAd_00018	GCTAACTTCCCCTATCCGCTTATAGGCAAGACCGCAGTTGACAGCATTACCCAGAAAAAGTTTCTTTGCG
PBAd_00019	ACAGTCCTTCCAACGTAAAAATTTCTGATAACCCAAACACCTACGACTACATGAACAAGCGAGTGGTGGC
PBAd_00020	AAGATGAACTTCCAAATTACTGCTTTCCACTGGGAGGTGTGATTAATACAGAGACTCTTACCAAGGTAAA
PBAd_00021	AGCTAACATGCTTTACCCCATCCCTGCCAATGCAACCAACATTCCAATTTCCATCCCATCTCGCAACTGG
PBAd_00022	TTCAACTCTTGAAGCCATGCTGCGCAACGATACCAATGATCAGTCATTCAACGACTACCTCTCTGCAGCT
PBAd_00023	AGGCTGTGGACAGCTATGATCCCGATGTTCGTATTATTGAAAATCATGGCGTCGAGGATGAACTGCCTAA
PBAd_00024	TGAAATTGTGCTTTACACGGAAAATGTCAATTTGGAAACTCCAGACAGCCATGTGGTATACAAGCCAGGA
PBAd_00025	CATCGGCTATCAGGGCTTCTACATTCCAGAAGGATACAAAGATCGCATGTATTCATTTTTCAGAAACTTC
PBAd_00026	GCTGCTTCTCCCAGGCTCCTACACTTATGAGTGGAACTTTAGGAAGGATGTGAACATGGTTCTACAGAGT
PBAd_00027	ATGACACCAATGATCAGTCATTCAACGACTACCTATCTGCAGCTAACATGCTCTACCCCATTCCTGCCAA
PBAd_00028	CTTGCCAACTACAACATTGGATACCAGGGCTTCTACGTTCCTGAGGGTTACAAGGATCGCATGTACTCCT
PBAd_00029	GATCGCATGTACTCCTTCTTCAGAAACTTCCAGCCCATGAGTAGACAGGTGGTTGATGAGATTAACTACA
PBAd_00030	CCCCTAAGGGCGCTCCCAATACATCTCAGTGGATTGCTGAAGGCGTAAAAAAAGAAGATGGGGGATCTGA
PBAd_00031	AGAAAATGTAAATTTGGAAACTCCAGATTCCCATGTTGTTTACAAAGCAGGAACTTCAGACGAAAGCTCT
PBAd_00032	TGTGGCTACCAATACTGTTTACCAAGGTGTTAAGTTACAAACTGGTCAAACTGACAAATGGCAGAAAGAT
PBAd_00033	CCGAATTGGGAAGGGTAGCGTATTCGCCATGGAAATCAATCTCCAGGCCAACCTGTGGAAGAGTTTTCTG
PBAd_00034	TTGATGAGGTCAATTACAAAGACTTCAAGGCCGTCGCCATACCCTACCAACACAACAACTCTGGCTTTGT
PBAd_00035	TGACGAAGAGGAAGAGAAAAATCTCACCACTTACACTTTTGGAAATGCCCCAGTGAAAGCAGAAGGTGGT
PBAd_00036	AGAAGATTTTGACATTGACATGGCTTTCTTTGATTCCAACACTATTAACACACCAGATGTTGTGCTGTAT

Table 11. Exemplary HCoV-OC229E Probes

Id	Sequence
PBS10049	AATGGGGTTATGTTGGTTCACTCTCCACTAATCACCATGCAATTTGTAATGTTCATAGAAATGAGCATGT
PBS10050	GTGTATGACTGCTTTGTTAAGAATGTGGATTGGTCAATTACCTACCCTATGATAGCTAATGAAAATGCCA
PBS10051	TTGCATCTTCTTTTGTTGGTATGCCATCTTTTGTTGCATATGAAACAGCAAGACAAGAGTATGAAAATGC
PBS10052	AAATGGTTCCTCACCACAAATAATCAAACAATTGAAGAAGGCTATGAATGTTGCAAAAGCTGAGTTTGAC
PBS10053	CTGCTGCAGCTATGTACAAAGAAGCACGTGCTGTTAATAGAAAATCAAAAGTTGTTAGTGCCATGCATAG
PBS10054	ACGTTTGGACATGTCTAGTGTTGACACTATCCTTAATATGGCACGTAATGGTGTTGTCCCTCTTTCCGTT
PBS10055	CTGGTGGTAAAGTTTCATTTTCTGATGACGTTGAAGTAAAAGACATTGAACCTGTTTACAGAGTCAAGCT
PBS10056	TTTACAGAGTCAAGCTTTGCTTTGAGTTTGAAGATGAAAAACTTGTAGATGTTTGTGAAAAGGCAATTGG
PBS10057	GATGTTTGTGAAAAGGCAATTGGCAAGAAAATTAAACATGAAGGTGACTGGGATAGCTTTTGTAAGACTA
PBS10058	GCGTTGTTGGCCTTTTTCTTGTCTAAGCATAGTGATTTTGGTCTTGGTGATCTTGTCGATTCTTATTTTG
PBS10059	AGCAAGACAAGAGTATGAAAATGCTGTTGCAAATGGTTCCTCACCACAAATAATCAAACAATTGAAGAAG
PBS10060	TTGAAGAAGGCTATGAATGTTGCAAAAGCTGAGTTTGACAGGGAATCATCTGTTCAAAAGAAAATTAACA
PBS10061	CTGCTGCAGCTATGTACAAAGAAGCACGTGCTGTTAATAGAAAATCAAAAGTTGTTAGTGCCATGCATAG

Table 12. Exemplary HCoV-OC43 Probes

Id	Sequence
PBS10062	CTCACATCCTAGGAAGATGCATAGTTTTAGATGTTAAAGGTGTAGAAGAATTGCATGACGATTTAGTTAA
PBS10063	GGATTGGCCATTGCACCATAGCTCAACTCACGGATGCAGCACTGTCCATTAAGGAAAATGTTGATTTTAT
PBS10064	GCATGCAATTCAATTATAAAATCACCATCAACCCCTCATCACCGGCTAGACTTGAAATAGTTAAGCTCGG
PBS10065	ATAGTTAGTCACTGGATGGGAATTCGTTTTGAATACACATCACCCACTGATAAGCTAGCTATGATTATGG

In yet another aspect, the present invention is directed to a kit for hybridization analysis of a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43, which kit comprises: a) a above-described probe; and b) a means for assessing a hybrid formed between a nucleotide sequence of an influenza A virus, an influenza B virus, a human metapneumovirus, a human adenovirus, a human coronaviruse 229E or a human coronaviruse OC43 and said probe.

BRIEF DESCRIPTION OF THE SEVERAL VIEWS OF THE DRAWING(S)

Figure 1A and 1B illustrate exemplary SARS-CoV genome structures (*See* Figure 2 of Marra et al., Science 2003 May 1; [epub ahead of print]; and GenBank Accession No. NC_004718).

Figure 2 illustrates an exemplary sample preparation procedure.

Figure 3 illustrates an exemplary probe labeling to be used in PCR. The sequence of the universal primer is complementary to the common sequence of the specific primer. The universal primers and the specific primers are added into the PCR master mix before the amplification are performed. The specificity of the amplification is ensured by the specific part of the specific primer. After one or a few thermal cycles, the universal primer can be incorporated into the amplicon efficiently. Then the universal primer can anneal to the complementary sequence of the common sequence of the specific primer The PCR can further proceed with the fluorescence dye incorporated in the universal primer. 1 and 6 depict a fluorescence dye; 2 depicts an upstream universal primer; 3 depicts an upstream specific primer with a common sequence; 4 depicts a template; 5 depicts a downstream specific primer with a common sequence; and 7 depicts a downstream universal primer.

Figure 4 illustrates probe immobilization on a glass slide surface modified with an amino group, *e.g.*, poly-L-lysine treated. Amine Coupling Chemistry: Amine Substrates contain primary amine groups ($NH3^+$) attached covalently to the glass surface (rectangles). The amines carry a positive charge at neutral pH, allowing attachment of natively charged DNA (double helix) through the formation of ionic bonds with the negatively charged phosphate backbone (middle panel). Electrostatic attachment is supplemented by treatment with an ultraviolet light or heat, which induces covalent attachment of the DNA to the surface through the covalent binding between the primary amine and thymine (right panel). The combination of electrostatic binding and covalent attachment couples the DNA to the substrate in a highly stable manner.

Figure 5 illustrates an exemplary array format of SARS-CoV detection chip.

Figure 6A and 6B illustrate SARS-CoV detection from a SARS patient blood sample (sample No. 3).

Figure 7A and 7B illustrate SARS-CoV detection from a SARS patient blood sample (sample No. 4).

Figure 8A and 8B illustrate SARS-CoV detection from a SARS patient sputum sample (sample No. 5).

Figure 9A and 9B illustrate SARS-CoV detection from a SARS patient sputum sample (sample No. 6).

Figure 10 illustrates another exemplary array format of SARS-CoV detection chip.

Figure 11 illustrates all possible positive results on the SARS SARS-CoV detection chip illustrated in Figure 10.

Figure 12 illustrates another exemplary array format of SARS-CoV detection chip.

Figure 13 illustrates all possible positive results on the SARS SARS-CoV detection chip illustrated in Figure 12.

Figure 14 illustrates all possible positive and negative results on the SARS SARS-CoV detection chip illustrated in Figure 12.

DETAILED DESCRIPTION OF THE INVENTION

For clarity of disclosure, and not by way of limitation, the detailed description of the invention is divided into the subsections that follow.

A. Definitions

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as is commonly understood by one of ordinary skill in the art to which this invention belongs. All patents, applications, published applications and other publications referred to herein are incorporated by reference in their entirety. If a definition set forth in this section is contrary to or otherwise inconsistent with a definition set forth in the patents, applications, published applications and other publications that are herein incorporated by reference, the definition set forth in this section prevails over the definition that is incorporated herein by reference.

As used herein, "a" or "an" means "at least one" or "one or more."

As used herein, "coronaviridae" refers to a family of single-stranded RNA viruses responsible for respiratory diseases. The outer envelope of the virus has club-shaped projections that radiate outwards and give a characteristic corona appearance to negatively stained virions.

As used herein, "polymerase chain reaction (PCR)" refers to a system for *in vitro* amplification of DNA. Two synthetic oligonucleotide primers, which are complementary to two regions of the target DNA (one for each strand) to be amplified, are added to the target DNA (that need not be pure), in the presence of excess deoxynucleotides and a heat-stable DNA polymerase, *e.g.*, Taq DNA polymerase. In a series, *e.g.*, 30, of temperature cycles, the target DNA is repeatedly denatured (*e.g.*, around 90°C), annealed to the primers (*e.g.*, at 50-60°C) and a daughter strand extended from the primers (*e.g.*, 72°C). As the daughter strands themselves act as templates for subsequent cycles, DNA fragments matching both primers are amplified exponentially, rather than linearly. The original DNA need thus be neither pure nor abundant, and the PCR reaction has accordingly become widely used not only in research, but in clinical diagnostics and forensic science.

As used herein, "nested PCR" refers to a PCR in which specificity is improved by using two sets of primers sequentially. An initial PCR is performed with the "outer" primer pairs, then a small aliquot is used as a template for a second round of PCR with the "inner" primer pair.

As used herein, "reverse transcription PCR or RT-PCR" refers to PCR in which the starting template is RNA, implying the need for an initial reverse transcriptase step to make a DNA template. Some thermostable polymerases have appreciable reverse transcriptase activity; however, it is more common to perform an explicit reverse transcription, inactivate the reverse transcriptase or purify the product, and proceed to a separate conventional PCR.

As used herein, "primer" refers to an oligonucleotide that hybridizes to a target sequence, typically to prime the nucleic acid in the amplification process.

As used herein, "probe" refers to an oligonucleotide that hybridizes to a target sequence, typically to facilitate its detection. The term "target sequence" refers to a nucleic acid sequence to which the probe specifically binds. Unlike a primer that is used to prime the target nucleic acid in the amplification process, a probe need not be extended to amplify target sequence using a polymerase enzyme. However, it will be apparent to those skilled in the art that probes and primers are structurally similar or identical in many cases.

As used herein, "the concentration of said 5' and 3' universal primers equals to or is higher than the concentration of said 5' and 3' specific primers, respectively" means that the concentration of the 5' universal primer equals to or is higher than the concentration of the 5' specific primers and the concentration of the 3' universal primer equals to or is higher than the concentration of the 3' specific primers.

As used herein, "hairpin structure" refers to a polynucleotide or nucleic acid that contains a double-stranded stem segment and a single-stranded loop segment wherein the two polynucleotide or nucleic acid strands that form the double-stranded stem segment is linked and separated by the single polynucleotide or nucleic acid strand that forms the loop segment. The "hairpin structure" can further comprise 3' and/or 5' single-stranded region(s) extending from the double-stranded stem segment.

As used herein, "nucleic acid (s)" refers to deoxyribonucleic acid (DNA) and/or ribonucleic acid (RNA) in any form, including *inter alia*, single-stranded, duplex, triplex, linear and circular forms. It also includes polynucleotides, oligonucleotides, chimeras of nucleic acids and analogues thereof. The nucleic acids described herein can be composed of the well-known deoxyribonucleotides and ribonucleotides composed of the bases adenosine, cytosine, guanine, thymidine, and uridine, or may be composed of analogues or derivatives of these bases. Additionally, various other oligonucleotide derivatives with nonconventional phosphodiester backbones are also included herein, such as phosphotriester, polynucleopeptides (PNA), methylphosphonate, phosphorothioate, polynucleotides primers, locked nucleic acid (LNA) and the like.

As used herein, "complementary or matched" means that two nucleic acid sequences have at least 50% sequence identity. Preferably, the two nucleic acid

sequences have at least 60%, 70,%, 80%, 90%, 95%, 96%, 97%, 98%, 99% or 100% of sequence identity. "Complementary or matched" also means that two nucleic acid sequences can hybridize under low, middle and/or high stringency condition(s).

As used herein, "substantially complementary or substantially matched" means that two nucleic acid sequences have at least 90% sequence identity. Preferably, the two nucleic acid sequences have at least 95%, 96%, 97%, 98%, 99% or 100% of sequence identity. Alternatively, "substantially complementary or substantially matched" means that two nucleic acid sequences can hybridize under high stringency condition(s).

As used herein, "two perfectly matched nucleotide sequences" refers to a nucleic acid duplex wherein the two nucleotide strands match according to the Watson-Crick basepair principle, *i.e.*, A-T and C-G pairs in DNA:DNA duplex and A-U and C-G pairs in DNA:RNA or RNA:RNA duplex, and there is no deletion or addition in each of the two strands.

As used herein: "stringency of hybridization" in determining percentage mismatch is as follows:

1) high stringency: 0.1 x SSPE (or 0.1 x SSC), 0.1% SDS, 65°C;

2) medium stringency: 0.2 x SSPE (or 1.0 x SSC), 0.1% SDS, 50°C (also referred to as moderate stringency); and

3) low stringency: 1.0 x SSPE (or 5.0 x SSC), 0.1% SDS, 50°C.

It is understood that equivalent stringencies may be achieved using alternative buffers, salts and temperatures.

As used herein, "gene" refers to the unit of inheritance that occupies a specific locus on a chromosome, the existence of which can be confirmed by the occurrence of different allelic forms. Given the occurrence of split genes, gene also encompasses the set of DNA sequences (exons) that are required to produce a single polypeptide.

As used herein, "melting temperature" ("Tm") refers to the midpoint of the temperature range over which nucleic acid duplex, *i.e.*, DNA:DNA, DNA:RNA, RNA:RNA, PNA: DNA, LNA:RNA and LNA: DNA, etc., is denatured.

As used herein, "sample" refers to anything which may contain a target SARS-CoV to be assayed or amplified by the present chips, primers, probes, kits and methods. The sample may be a biological sample, such as a biological fluid or a biological tissue. Examples of biological fluids include urine, blood, plasma, serum, saliva, semen, stool, sputum, cerebral spinal fluid, tears, mucus, amniotic fluid or the like. Biological tissues are aggregates of cells, usually of a particular kind together with their intercellular substance that form one of the structural materials of a human, animal, plant, bacterial, fungal or viral structure, including connective, epithelium, muscle and nerve tissues. Examples of biological tissues also include organs, tumors, lymph nodes, arteries and individual cell(s). Biological tissues may be processed to obtain cell suspension samples. The sample may also be a mixture of cells prepared *in vitro*. The sample may also be a cultured cell suspension. In case of the biological samples, the sample may be crude samples or processed samples that are obtained after various processing or preparation on the original samples. For example, various cell separation methods (*e.g.*, magnetically activated cell sorting) may be applied to separate or enrich target cells from a body fluid sample such as blood. Samples used for the present invention include such target-cell enriched cell preparation.

As used herein, a "liquid (fluid) sample" refers to a sample that naturally exists as a liquid or fluid, *e.g.*, a biological fluid. A "liquid sample" also refers to a sample that naturally exists in a non-liquid status, *e.g.*, solid or gas, but is prepared as a liquid, fluid, solution or suspension containing the solid or gas sample material. For example, a liquid sample can encompass a liquid, fluid, solution or suspension containing a biological tissue.

As used herein, "assessing PCR products" refers to quantitative and/or qualitative determination of the PCR products, and also of obtaining an index, ratio, percentage, visual or other value indicative of the level of the PCR products. Assessment may be direct or indirect and the chemical species actually detected need not of course be the PCR products themselves but may, for example, be a derivative thereof, or some further substance.

B. Chips for assaying for a SARS-CoV and a non-SARS-CoV infectious organism

In one aspect, the present invention is directed to a chip for assaying for a coronaviruse causing the severe acute respiratory syndrome (SARS-CoV) and a non-SARS-CoV infectious organism, which chip comprises a support suitable for use in nucleic acid hybridization having immobilized thereon an oligonucleotide probe complementary to a nucleotide sequence of SARS-CoV genome, said nucleotide sequence comprising at least 10 nucleotides, and one or more of the following oligonucleotide probe(s): a) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism causing SARS-like symptoms, said nucleotide sequence comprising at least 10 nucleotides; b) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism damaging an infectious host's immune system, said nucleotide sequence comprising at least 10 nucleotides; or c) an oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV coronaviridae virus, said nucleotide sequence comprising at least 10 nucleotides.

In some embodiments, the chip comprises a support suitable for use in nucleic acid hybridization having immobilized thereon at least two oligonucleotide probes complementary to at least two different nucleotide sequences of SARS-CoV genome, each of said two different nucleotide sequences comprising at least 10 nucleotides.

The at least two different nucleotide sequences can be any suitable combinations. For example, the at least two different nucleotide sequences of SARS-CoV genome can comprise a nucleotide sequence of at least 10 nucleotides located within a conserved region of SARS-CoV genome and a nucleotide sequence of at least 10 nucleotides located within a variable region of SARS-CoV genome. In another example, the at least two different nucleotide sequences of SARS-CoV genome can comprise a nucleotide sequence of at least 10 nucleotides located within a structural protein coding gene of SARS-CoV genome and a nucleotide sequence of at least 10 nucleotides located within a non-structural protein coding gene of SARS-CoV genome.

If desired, the present chips can comprise other types of probes or other features. For example, the chip can further comprise: a) at least one of the following three oligonucleotide probes: an immobilization control probe that is labeled and does not

participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV or a non-SARS-CoV infectious organism is contacted with the chip, a positive control probe that is not complementary to any SARS-CoV or non-SARS-CoV infectious organism sequence but is complementary to a sequence contained in the sample not found in the SARS-CoV or the non-SARS-CoV infectious organism and a negative control probe that is not complementary to any nucleotide sequence contained in the sample; and b) a blank spot.

In a specific embodiment, the present chips can comprise at least two oligonucleotide probes complementary to two different nucleotide sequences of at least 10 nucleotides, respectively, located within a conserved region of SARS-CoV genome, located within a structural protein coding gene of SARS-CoV genome or located within a non-structural protein coding gene of SARS-CoV genome.

Any conserved region of SARS-CoV genome can be used as assay target. For example, the conserved region of SARS-CoV genome can be a region located within the Replicase 1A, 1B gene or the Nucleocapsid (N) gene of SARS-CoV.

Any variable region of SARS-CoV genome can be used as assay target. For example, the variable region of SARS-CoV genome can be a region located within the Spike glycoprotein (S) gene of SARS-CoV.

Any structural protein coding gene of SARS-CoV genome can be used as assay target. For example, the structural protein coding gene of SARS-CoV genome can be a gene encoding the Spike glycoprotein (S), the small envelope protein (E) or the Nucleocapsid protein (N).

Any non-structural protein coding gene of SARS-CoV genome can be used as assay target. For example, the non-structural protein coding gene of SARS-CoV genome can be a gene encoding the Replicase 1A or 1B.

In another specific embodiment, the present chips can comprise at least two of the following four oligonucleotide probes: two oligonucleotide probes complementary to two different nucleotide sequences of at least 10 nucleotides located within the Replicase 1A or 1B gene of SARS-CoV, an oligonucleotide probe complementary to a nucleotide sequence of at least 10 nucleotides located within the N gene of SARS-CoV and an

oligonucleotide probe complementary to a nucleotide sequence of at least 10 nucleotides located within the S gene of SARS-CoV.

Preferably, one or both of the different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV can comprise a nucleotide sequence that: a) hybridizes, under high stringency, with a Replicase 1A or 1B nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13; or b) has at least 90% identity to a Replicase 1A or 1B nucleotide sequence comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13. More preferably, one or both of the different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV comprises a nucleotide sequence that is set forth in Table 13.

Table 13. Exemplary SARS-CoV probes

probe_id	Sequence 5' -3'	region
PBS00001	TTACCCTAATATGTTTATCACCCGCGAAGAAGCTATTCGTCACGTTCGTGCGTGGA	SARS-Cov Replicase 1B
PBS00002	CTGACAAGTATGTCCGCAATCTACAACACAGGCTCTATGAGTGTCTCTATAGAAAT	SARS-Cov Replicase 1B
PBS00003	CATAACACTTGCTGTAACTTATCACACCGTTTCTACAGGTTAGCTAACGAGTGTGC	SARS-Cov Replicase 1B
PBS00004	TTACCCTAATATGTTTATCACCCGCGAAGAAGCTATTCGTCACGTTCGTG	SARS-Cov Replicase 1B
PBS00009	GCGTTCTCTTAAAGCTCCTGCCGTAGTGTCAGTATCATCACCAGATGCTGTTACTACATATAATGGATAC	SARS-Cov Replicase 1A
PBS00010	CTTTGGCTGGCTCTTACAGAGATTGGTCCTATTCAGGACAGCGTACAGAGTTAGGTGTTGAATTTCTTAA	SARS-Cov Replicase 1A
PBS00011	CTACGTAGTGAAGCTTTCGAGTACTACCATACTCTTGATGAGAGTTTTCTTGGTAGGTACATGTCTGCTT	SARS-Cov Replicase 1A
PBS00012	TGCCAATTGGTTATGTGACACATGGTTTTAATCTTGAAGAGGCTGCGCCGCTGTATGCGTTCTCTTAAAGC	SARS-Cov Replicase 1A
PBS00013	TATAAAGTTACCAAGGGAAAGCCCGTAAAAGGTGCTTGGAACATTGGACAACAGAGATCAGTTTTAACAC	SARS-Cov Replicase 1A
PBS00014	TGCTTCATTGATGTTGTTAACAAGGCACTCGAAATGTGCATTGATCAAGTCACTATCGCTGGCGCAAAG	SARS-Cov Replicase 1A
PBS00015	TGTCGACGCCATGGTTTATACTTCAGACCTGCTCACCAACAGTGTCATTATTATGGCATATGTAACTGGT	SARS-Cov Replicase 1A
PBS00016	TACTGTTGAAAAACTCAGGCCTATCTTTGAATGGATTGAGGCGAAACTTAGTGCAGGAGTTGAATTTCTC	SARS-Cov Replicase 1A
PBS00017	ACCTATTCTGTTGCTTGACCAAGCTCTTGTATCAGACGTTGGAGATAGTACTGAAGTTTCC	SARS-Cov Replicase 1A
PBS00018	GCCTATTAATGTCATAGTTTTTGATGGCAAGTCCAAATGCGACGAGTCTGCTTCTAAGTCTGCTTCTGTG	SARS-Cov Replicase 1A
PBS00019	TGAGAGCTAACAACACTAAAGGTTCACTGCCTATTAATGTCATAGTTTTTGATGGCAAGTCCAAATGCGA	SARS-Cov Replicase 1A
PBS00020	ACTTGCATGATGTGCTATAAGCGCAATCGTGCCACACGCGTTGAGTGTACAACTATTGTTAATGGCATGA	SARS-Cov Replicase 1A
PBS00021	GGCGATGTAGTGGCTATTGACTATAGACACTATTCAGCGAGTTTCAAGAAAGGTGCTAAATTACTGCATA	SARS-Cov Replicase 1A

PBS00022	TCAAACCAAACACTTGGTGTTTACGTTGTCTTTGGAGTACAAAGCCAGTAGATACTTCAAATTCATTTGA	SARS-Cov Replicase 1A
PBS00023	TAGTGCTGTTGGCAACATTTGCTACACACCTTCCAAACTCATTGAGTATAGTGATTTTGCTAC	SARS-Cov Replicase 1A
PBS00024	TCATAGCTAACATCTTTACTCCTCTTGTGCAACCTGTGGGTGCTTTAGATGTGTCTGCTTCAGTAGTGGC	SARS-Cov Replicase 1A
PBS00025	GGTATTATTGCCATATTGGTGACTTGTGCTGCCTACTACTTTATGAAATTCAGACGTGTTTTTGGTGAGT	SARS-Cov Replicase 1A
PBS00026	GTGATGTCAGAGAAACTATGACCCATCTTCTACAGCATGCTAATTTGGAATCTGCAAAGCGAGTTCTTAA	SARS-Cov Replicase 1A
PBS00027	AACCATCAAGCCTGTGTCGTATAAACTCGATGGAGTTACTTACACAGAGATTGAACCAAAATTGGATGGG	SARS-Cov Replicase 1A
PBS00028	GTTTTCTACAAGGAAACATCTTACACTACAACCATCAAGCCTGTGTCGTATAAACTCGATGGAGTTACTT	SARS-Cov Replicase 1A
PBS00029	CCTTGAATGAGGATCTCCTTGAGATACTGAGTCGTGAACGTGTTAACATTAACATTGTTGGCGATTTTCA	SARS-Cov Replicase 1A
PBS00031	GCCATGGTTTATACTTCAGACCTGCTCACCAACAGTGTCATTATTATGGCATATGTAACTGGTGGTCTTG	SARS-Cov Replicase 1A
PBS00032	CAACAGACTTCTCAGTGGTTGTCTAATCTTTTGGGCACTACTGTTGAAAAACTCAGGCCTATCTTTGAAT	SARS-Cov Replicase 1A
PBS00033	TTCCCGTCAGGCAAAGTTGAAGGGTGCATGGTACAAGTAACCTGTGGAACTACAAC	SARS-Cov Replicase 1A
PBS00034	GGTTCACCATCTGGTGTTTATCAGTGTGCCATGAGACCTAATCATACCATTAAAGG	SARS-Cov Replicase 1A
PBS00035	AGATCATGTTGACATATTGGGACCTCTTTCTGCTCAAACAGGAATTGCCGTC	SARS-Cov Replicase 1A
PBS00036	TAAAAAGGACAAAAAGAAAAAGACTGATGAAGCTCAGCCTTTGCCCGCAGAGACAAAAGAAGCAGCCCACT	SARS-Cov Nucleocapsid gene
PBS00037	ACGGCAAAATGAAAGAGCTCAGCCCCAGATGGTACTTCTATTACCTAGGAACTGGCCCAGAAGCTTCACT	SARS-Cov Nucleocapsid gene
PBS00038	GGCGCTAACAAAGAAGGCATCGTATGGGTTGCAACTGAGGGAGCCTTGAATACACCCAAAGACCACATTG	SARS-Cov Nucleocapsid gene
PBS00039	GTCCAGATGACCAAATTGGCTACTACCGAAGAGCTACCCGACGAGTTCGTGGTGGTGACGGCAAAATGAA	SARS-Cov Nucleocapsid gene
PBS00040	GAGGTCGTGAAACTGCCCTCGCGCTATTGCTGCTAGACAGATTGAACCAGCTTGAGAGCAAAGTTTCTGG	SARS-Cov Nucleocapsid gene
PBS00041	AAAAGAAAAAGACTGATGAAGCTCAGCCTTTGCCGCAGAGACAAAAGAAGCAGCCCACTGTGACTCTTCT	SARS-Cov Nucleocapsid gene
PBS00042	AAATTGCACAATTTGCTCCAAGTGCCTCTGCATTCTTTGGAATGTCACGCATTGGCATGGAAGTCACACC	SARS-Cov Nucleocapsid gene
PBS00043	ACCAATTTAACAAGGCGATTAGTCAAATTCAAGAATCACTTACAACAACATCAACTGCATTGGGCAAGCT	SARS-Cov Spike glycoprotein gene
PBS00044	CACCTGGAACAAATGCTTCATCTGAAGTTGCTGTTCTATATCAAGATGTTAACTGCACTGATGTTTCTAC	SARS-Cov Spike glycoprotein gene
PBS00045	AAAGGGCTACCACCTTATGTCCTTCCCACAAGCAGCCCCGCATGGTGTTGTCTTCCTACATGTCACGTAT	SARS-Cov Spike glycoprotein gene
PBS00046	TCAGGAAATTGTGATGTCGTTATTGGCATCATTAACAACACAGTTTATGATCCTCTGCAACCTGAGCTTG	SARS-Cov Spike glycoprotein gene
PBS00047	TTGATCTTGGCGACATTTCAGGCATTAACGCTTCTGTCGTCAACATTCAAAAAGAAATTGACCGCCTCAA	SARS-Cov Spike glycoprotein gene
PBS00048	GAGGAACTTCACCACAGCGCCAGCAATTTGTCATGAAGGCAAAGCATACTTCCCTCGTGAAGGTGTTTTT	SARS-Cov Spike glycoprotein gene

Also preferably, the nucleotide sequence located within the N gene of SARS-CoV can comprise a nucleotide sequence that: a) hybridizes, under high stringency, with a N nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13; or b)

has at least 90% identity to a N nucleotide sequence comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13. More preferably, the nucleotide sequence located within the N gene of SARS-CoV comprises a nucleotide sequence that is set forth in Table 13.

Also preferably, the nucleotide sequence located within the S gene of SARS-CoV can comprise a nucleotide sequence that: a) hybridizes, under high stringency, with a S nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13; or b) has at least 90% identity to a S nucleotide sequence comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13. More preferably, the nucleotide sequence located within the S gene of SARS-CoV comprises a nucleotide sequence that is set forth in Table 13.

Any suitable label can be used in the immobilization control probe, *e.g.*, a chemical, an enzymatic, an immunogenic, a radioactive, a fluorescent, a luminescent or a FRET label.

Any suitable non-SARS-CoV-sequence can be used. For example, the non-SARS-CoV-sequence can be an endogenous component of a sample to be assayed. Alternatively, the non-SARS-CoV-sequence is spiked in the sample to be assayed. In another example, the spiked non-SARS-CoV-sequence can be a sequence of *Arabidopsis* origin.

In still another specific embodiment, the present chips can comprise two oligonucleotide probes complementary to two different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV, an oligonucleotide probe complementary to a nucleotide sequence located within the N gene of SARS-CoV, an oligonucleotide probe complementary to a nucleotide sequence located within the S gene of SARS-CoV, an immobilization control probe that is labeled and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV or a non-SARS-CoV infectious organism is contacted with the chip, a positive control probe that is not complementary to any SARS-CoV sequence but is complementary to any sequence contained in the sample not found in the SARS-CoV or

the non-SARS-CoV infectious organism and a negative control probe that is not complementary to any nucleotide sequence contained in the sample.

Preferably, the chip comprises multiple spots of the described probes, *e.g.*, multiple spots of the two oligonucleotide probes complementary to two different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV, the oligonucleotide probe complementary to a nucleotide sequence located within the N gene of SARS-CoV, the oligonucleotide probe complementary to a nucleotide sequence located within the S gene of SARS-CoV, the immobilization control probe, the positive control probe and the negative control probe.

The present chips can further comprise an oligonucleotide probe complementary to a nucleotide sequence of a coronaviruse not related to the SARS-CoV. For example, the coronaviruse not related to the SARS can be the Group I, II or III coronaviruse or is a coronaviruse that infects an avian species, *e.g.*, Avian infectious bronchitis virus and Avian infectious laryngotracheitis virus, an equine species, *e.g.*, Equine coronaviruse, a canine species, *e.g.*, Canine coronaviruse, a feline species, *e.g.*, Feline coronaviruse and Feline infectious peritonitis virus, a porcine species, *e.g.*, Porcine epidemic diarrhea virus, Porcine transmissible gastroenteritis virus and Porcine hemagglutinating encephalomyelitis virus, a calf species, *e.g.*, Neonatal calf diarrhea coronaviruse, a bovine species, *e.g.*, Bovine coronaviruse, a murine species, *e.g.*, Murine hepatitis virus, a puffinosis species, *e.g.*, Puffinosis virus, a rat species, *e.g.*, Rat coronaviruse and a Sialodacryoadenitis virus of rat, *e.g.*, a turkey species *e.g.*, Turkey coronaviruse, or a human species, *e.g.*, Human enteric coronaviruse. The present chips can further comprise an oligonucleotide probe complementary to a nucleotide sequence of other types of virus or pathogens. An exemplary list of viruses and pathogens that can be assayed using the present chips is set forth in the following Table 14.

Table 14. Exemplary viruses and pathogens

No.	Virus name	Genome	Sample nucleic acid	Structure
1	Coronaviridae	Single-stranded,	RNA	Having capsid

		linear RNA		
2	SARS-CoV	Single-stranded, linear RNA	RNA	Having capsid
3	Human coronaviruse 229E	Single-stranded, linear RNA	RNA	Having capsid
4	Human coronaviruse OC43	Single-stranded, linear RNA	RNA	Having capsid
5	Influenzavirus A,B,C	Single-stranded, linear RNA, fragmented	RNA	Having capsid
6	Parainfluenza virus	Single-stranded, linear RNA	RNA	Having capsid
7	Respiratory sncytical virus	Single-stranded, linear RNA	RNA	Having capsid
8	Human metapneumovirus	Single-stranded, linear RNA	RNA	Having capsid
9	Rhinovirus	Single-stranded RNA	RNA	No capsid
10	Adenoviruse	Double-stranded, linear DNA	DNA	No capsid
11	Mycoplasma pneumoniae	Double-stranded, linear DNA	DNA and RNA	Having cell wall
12	Chlamydia pneumoniae	Double-stranded, linear DNA	DNA and RNA	No cell wall

The various probes, *e.g.*, the oligonucleotide probe complementary to a nucleotide sequence located within a conserved region of SARS-CoV genome, the oligonucleotide probe complementary to a nucleotide sequence located within a variable

region of SARS-CoV genome, the immobilization control probe, the positive control probe or the negative control probe the oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism causing SARS-like symptoms, the oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV infectious organism damaging an infectious host's immune system, and the oligonucleotide probe complementary to a nucleotide sequence of a non-SARS-CoV coronaviridae virus, can comprise, at its '5 end, a poly dT region to enhance its immobilization on the support.

In a specific embodiment, the at least one of the oligonucleotide probes is complementary to a highly expressed nucleotide sequence of SARS-CoV genome. Such a chip is particularly useful in detecting early-stage SARS-CoV infection.

In some embodiments, the non-SARS-CoV infectious organism is an infectious organism causing SARS-like symptoms. Such organism includes, but not limited to, a human coronaviruse 229E, a human coronaviruse OC43, a human enteric coronaviruse, an influenza virus, a parainfluenza virus, a respiratory sncytical virus, a human metapneumovirus, a rhinovirus, an adenoviruse, a mycoplasma pneumoniae, a chlamydia pneumoniae, a measles virus and a rubella virus. The influenza virus can be influenza virus A or influenza virus B. The parainfluenza virus can be parainfluenza virus 1, parainfluenza virus 2, parainfluenza virus 3, or parainfluenza virus 4. Exemplary probes for these organisms are set forth in Table 15.

Table 15. Exemplary probes for non-SARS-CoV infectious organisms causing SARS-like symptoms

seqId	sequence(5'-3')	species
PBIA_00001	TTTAGAGCCTATGTGGATGGATTCRAACCGAACGGCTGCATTGAGGG CAAGCTTTCTCAAATGTC	Influenza A virus
PBIA_00002	ACAATTGAAGAAAGATTTGAAATCACTGGAACCATGCGCAGGCTTGCC GACCAAAGTCTCCCACCGAACT	Influenza A virus
PBIA_00003	AGCAATNGAGGAGTGCCTGATTAANGATCCCTGGGTTTTGCTNAATG	Influenza A virus

	C	
PBIA_00004	CCATACAGCCATGGAACAGGAACAGGATACACCATGGACACAGTCAA CAGAACACANCAATATTCAGAAA	Influenza A virus
PBIA_00005	GGGCGGGGAGTCTTCGAGCTCTCNGACGAAAAGGCAACGAACCCGA TCGTGCC	Influenza A virus
PBIA_00006	GATCTNGAGGCTCTCATGGAATGGCTAAAGACAAGACCAATCCTGTC ACCTCTGACTAA	Influenza A virus
PBIB_00001	GCTGGGAAATAGCATGGAACTGATGATATTCAGCTACAATCAAGACTA TTCGTTAAGTAATGAATCCTCA	Influenza B virus
PBIB_00002	TCTGTTCCAGCTGGTTTCTCCAATTTTGAAGGAATGAGGAGCTACATA GACAATATAGATCCTAAAGGAG	Influenza B virus
PBIB_00003	TTACAACCATGAGCTACCAGAAGTTCCATATAATGCCTTTCTTCTAATG TCTGATGAATTGGGGCTGGCC	Influenza B virus
PBIB_00004	ACAAATAAGATCCAAATGAAATGGGGAATGGAAGCTAGAAGATGTCTG CTTCAATCAATGCAACAAATGG	Influenza B virus
PBIB_00005	GAGGGAATGTATTCTGGAATAGANGAATGTATTAGTAACAACCCTTGG GTAATACAGAGTGCATACTGGT	Influenza B virus
PBIB_00006	CTACCGTGTTGGGAGTAGCCGCACTAGGTATCAAAAACATTGGAAAC AAAGAATACTTATGGGATGGACT	Influenza B virus
PBIB_00007	GGCTATGACTGAAAGAATAACCAGAGACAGCCCAATTTGGTTCCGGG ATTTTTGTAGTATAGCACCGGTC	Influenza B virus
PBIB_00008	ACTGATCAGAGGAACATGATTCTTGAGGAACAATGCTACGCTAAGTGT TGCAACCTTTTTGAGGCCTGTT	Influenza B virus
PBIB_00009	AAAATCCCTTTGTNGGACATTTGTCTATTGAGGGCATCAAAGANGCAG ATATAACCCCAGCACATGGTCC	Influenza B virus
PBIB_00010	CTTGGAATACAAGGGAATACAACTTAAAACAAATGCTGAAGACATAGG AACCAAAGGCCAAATGTGCTCA	Influenza B virus
PBIB_00011	GTGGCAGGAGCAACATCAGCTGAGTTCATAGAAATGCTACACTGCTT ACAAGGTGAAAATTGGAGACAAA	Influenza B virus
PBIB_00012	GGAACCCATCCCCGGAAAGAGCAACCACAAGCAGTGAAGCTGATGTC	Influenza B virus

	GGAAGGAAAACCCAAAAGAAACA	
PBIB_00013	CTGTTTCCAAAGATCAAAGGCACTAAAAAGAGTTGGACTTGACCCTTC ATTAATCAGTACCTTTGCAGGA	Influenza B virus
PBIB_00014	AGAGTTTTGTCTGCATTAACAGGCACAGAATTCAAGCCTAGATCAGCA TTAAAATGCAAGGGTTTCCATG	Influenza B virus
PBIB_00015	GAGGGACGTGATGCAGATGTCAAAGGAAATCTACTCAAGATGATGAA TGACTCAATGGCTAAGAAAACCA	Influenza B virus
PBIB_00016	CCTATCAGGAATGGGAACAACAGCAACAAAAAAGAAAGGCCTGATTC TAGCTGAGAGAAAAATGAGAAGA	Influenza B virus
PBIB_00017	GCAAGTCAAAAGAATGGGGAAGGAATTGCAAAGGATGTAATGGAAGT GCTAAAGCAGAGCTCTATGGGAA	Influenza B virus
PBAd_00001	CTGACACCTACCAAGGTATAAAATCAAACGGAAACGGTAATCCTCAAA ACTGGACCAAAAATGACGATTT	Human adenovirus
PBAd_00002	TCCTCTACTCCAACATTGCACTGTACCTGCCTGACAAGCTAAAATACA CTCCTACAAATGTGGAAATATC	Human adenovirus
PBAd_00003	GCTATCGGAGGCAGAGTACTAAAAAAGACTACTCCCATGAAACCATG CTACGGATCGTATGCCAGACCTA	Human adenovirus
PBAd_00004	AGTATTGTTTTGTACAGTGAGGATGTTAATATGGAAACTCCTGATACTC ACATTTCATACAAACCAAGCA	Human adenovirus
PBAd_00005	GGGAAACGATCTTAGAGTTGACGGGGCTAGCATTAAGTTTGACAGCA TTTGTCTTTACGCCACCTTCTTC	Human adenovirus
PBAd_00006	TTGCCATTAAAAACCTCCTCCTCCTGCCAGGCTCATATACATATGAAT GGAACTTCAGGAAGGATGTTAA	Human adenovirus
PBAd_00007	TTGCAACACGTAATGAAATAGGAGTGGGTAACAACTTTGCCATGGAAA TTAACCTAAATGCCAACCTATG	Human adenovirus
PBAd_00008	TTGGGGTAACTGACACCTATCAAGCTATTAAGGCTAATGGCAATGGCT CAGGCGATAATGGAGATATTAC	Human adenovirus
PBAd_00009	AGGTATCAAGGCATTAAAGTTAAAACCGATGACGCTAATGGATGGGAA AAAGATGCTAATGTTGATACAG	Human adenovirus
PBAd_00010	GAGAAGTTTTCTGTACTCCAATGTGGCTTTGTACCTTCCAGATGTTTA	Human adenovirus

	CAAGTACACGCCACCTAACATT	
PBAd_00011	ATCAGTCATTTAACGACTACCTCTCTGCAGCTAACATGCTTTACCCCA TTCCTGCCAATGCAACCAACAT	Human adenovirus
PBAd_00012	CTACTTCGTATATTCTGGATCTATTCCCTACCTGGATGGCACCTTTTAC CTTAACCACACTTTCAAGAAG	Human adenovirus
PBAd_00013	ACCTGCCAGTGGAAGGATGCTAACAGCAAAATGCATACCTTTGGGGT AGCTGCCATGCCAGGTGTTACTG	Human adenovirus
PBAd_00014	ATAGAAGCTGATGGGCTGCCTATTAGAATAGATTCAACTTCTGGAACT GACACAGTAATTTATGCTGATA	Human adenovirus
PBAd_00015	TTGAAATTAAGCGCACCGTGGACGGCGAGGGGTACAACGTGGCCCA GTGCAACATGACCAAGGACTGGTT	Human adenovirus
PBAd_00016	CGGCAACGACCGGCTCCTGACGCCCAACGAGTTTGAAATTAAGCGCA CCGTGGACGGCGAGGGGTACAAC	Human adenovirus
PBAd_00017	CTCCAGTAACTTTATGTCCATGGGCGCACTCACAGACCTGGGCCAAA ACCTTCTCTACGCCAACTCCGCC	Human adenovirus
PBAd_00018	GCTAACTTCCCCTATCCGCTTATAGGCAAGACCGCAGTTGACAGCATT ACCCAGAAAAAGTTTCTTTGCG	Human adenovirus
PBAd_00019	ACAGTCCTTCCAACGTAAAAATTTCTGATAACCCAAACACCTACGACT ACATGAACAAGCGAGTGGTGGC	Human adenovirus
PBAd_00020	AAGATGAACTTCCAAATTACTGCTTTCCACTGGGAGGTGTGATTAATA CAGAGACTCTTACCAAGGTAAA	Human adenovirus
PBAd_00021	AGCTAACATGCTTTACCCCATCCCTGCCAATGCAACCAACATTCCAAT TTCCATCCCATCTCGCAACTGG	Human adenovirus
PBAd_00022	TTCAACTCTTGAAGCCATGCTGCGCAACGATACCAATGATCAGTCATT CAACGACTACCTCTCTGCAGCT	Human adenovirus
PBAd_00023	AGGCTGTGGACAGCTATGATCCCGATGTTCGTATTATTGAAAATCATG GCGTCGAGGATGAACTGCCTAA	Human adenovirus
PBAd_00024	TGAAATTGTGCTTTACACGGAAAATGTCAATTTGGAAACTCCAGACAG CCATGTGGTATACAAGCCAGGA	Human adenovirus
PBAd_00025	CATCGGCTATCAGGGCTTCTACATTCCAGAAGGATACAAAGATCGCAT	Human adenovirus

	GTATTCATTTTTCAGAAACTTC	
PBAd_00026	GCTGCTTCTCCCAGGCTCCTACACTTATGAGTGGAACTTTAGGAAGG ATGTGAACATGGTTCTACAGAGT	Human adenovirus
PBAd_00027	ATGACACCAATGATCAGTCATTCAACGACTACCTATCTGCAGCTAACA TGCTCTACCCCATTCCTGCCAA	Human adenovirus
PBAd_00028	CTTGCCAACTACAACATTGGATACCAGGGCTTCTACGTTCCTGAGGGT TACAAGGATCGCATGTACTCCT	Human adenovirus
PBAd_00029	GATCGCATGTACTCCTTCTTCAGAAACTTCCAGCCCATGAGTAGACAG GTGGTTGATGAGATTAACTACA	Human adenovirus
PBAd_00030	CCCCTAAGGGCGCTCCCAATACATCTCAGTGGATTGCTGAAGGCGTA AAAAAAGAAGATGGGGGATCTGA	Human adenovirus
PBAd_00031	AGAAAATGTAAATTTGGAAACTCCAGATTCCCATGTTGTTTACAAAGCA GGAACTTCAGACGAAAGCTCT	Human adenovirus
PBAd_00032	TGTGGCTACCAATACTGTTTACCAAGGTGTTAAGTTACAAACTGGTCA AACTGACAAATGGCAGAAAGAT	Human adenovirus
PBAd_00033	CCGAATTGGGAAGGGTAGCGTATTCGCCATGGAAATCAATCTCCAGG CCAACCTGTGGAAGAGTTTTCTG	Human adenovirus
PBAd_00034	TTGATGAGGTCAATTACAAAGACTTCAAGGCCGTCGCCATACCCTACC AACACAACAACTCTGGCTTTGT	Human adenovirus
PBAd_00035	TGACGAAGAGGAAGAGAAAAATCTCACCACTTACACTTTTGGAAATGC CCCAGTGAAAGCAGAAGGTGGT	Human adenovirus
PBAd_00036	AGAAGATTTTGACATTGACATGGCTTTCTTTGATTCCAACACTATTAAC ACACCAGATGTTGTGCTGTAT	Human adenovirus
PBS10062	CTCACATCCTAGGAAGATGCATAGTTTTAGATGTTAAAGGTGTAGAAG AATTGCATGACGATTTAGTTAA	HCoV-OC43
PBS10063	GGATTGGCCATTGCACCATAGCTCAACTCACGGATGCAGCACTGTCC ATTAAGGAAAATGTTGATTTTAT	HCoV-OC43
PBS10064	GCATGCAATTCAATTATAAAATCACCATCAACCCCTCATCACCGGCTA GACTTGAAATAGTTAAGCTCGG	HCoV-OC43
PBS10065	ATAGTTAGTCACTGGATGGGAATTCGTTTTGAATACACATCACCCACT	HCoV-OC43

	GATAAGCTAGCTATGATTATGG	
PBS10049	AATGGGGTTATGTTGGTTCACTCTCCACTAATCACCATGCAATTTGTA ATGTTCATAGAAATGAGCATGT	HCoV-229E
PBS10050	GTGTATGACTGCTTTGTTAAGAATGTGGATTGGTCAATTACCTACCCT ATGATAGCTAATGAAAATGCCA	HCoV-229E
PBS10051	TTGCATCTTCTTTTGTTGGTATGCCATCTTTTGTTGCATATGAAACAGC AAGACAAGAGTATGAAAATGC	HCoV-229E
PBS10052	AAATGGTTCCTCACCACAAATAATCAAACAATTGAAGAAGGCTATGAA TGTTGCAAAAGCTGAGTTTGAC	HCoV-229E
PBS10053	CTGCTGCAGCTATGTACAAAGAAGCACGTGCTGTTAATAGAAAATCAA AAGTTGTTAGTGCCATGCATAG	HCoV-229E
PBS10054	ACGTTTGGACATGTCTAGTGTTGACACTATCCTTAATATGGCACGTAA TGGTGTTGTCCCTCTTTCCGTT	HCoV-229E
PBS10055	CTGGTGGTAAAGTTTCATTTTCTGATGACGTTGAAGTAAAAGACATTG AACCTGTTTACAGAGTCAAGCT	HCoV-229E
PBS10056	TTTACAGAGTCAAGCTTTGCTTTGAGTTTGAAGATGAAAAACTTGTAGA TGTTTGTGAAAAGGCAATTGG	HCoV-229E
PBS10057	GATGTTTGTGAAAAGGCAATTGGCAAGAAAATTAAACATGAAGGTGAC TGGGATAGCTTTTGTAAGACTA	HCoV-229E
PBS10058	GCGTTGTTGGCCTTTTTCTTGTCTAAGCATAGTGATTTTGGTCTTGGT GATCTTGTCGATTCTTATTTTG	HCoV-229E
PBS10059	AGCAAGACAAGAGTATGAAAATGCTGTTGCAAATGGTTCCTCACCACA AATAATCAAACAATTGAAGAAG	HCoV-229E
PBS10060	TTGAAGAAGGCTATGAATGTTGCAAAAGCTGAGTTTGACAGGGAATCA TCTGTTCAAAAGAAAATTAACA	HCoV-229E
PBS10061	CTGCTGCAGCTATGTACAAAGAAGCACGTGCTGTTAATAGAAAATCAA AAGTTGTTAGTGCCATGCATAG	HCoV-229E
PBHE_00001	CGGGATAAGGCACTCTCTATCAGAATGGATGTCTTGCTGCTATAATAG ATAGAGAAGGTTATAGCAGACT	Human enteric coronaviruse
PBHE_00002	CCCTCGCAGGAAAGTCGGGATAAGGCACTCTCTATCAGAATGGATGT	Human enteric

	CTTGCTGCTATAATAGATAGAGA	coronaviruse
PBHE_00003	ATGGATGTTTGAGGACGCAGAGGAGAAGTTGGACAACCCTAGTAGTT CAGAGGTGGATATAGTATGCT	Human enteric coronaviruse
PBHE_00004	CCTTGGGTTATGTACTTGCGTAAGTGTGGCGAAAAGGGTGCCTACAA TAAAGATCATAAACGTGTCGG	Human enteric coronaviruse
PBHE_00005	GGGGATGCTGGTTTTACTAGCATACTCAGTGGTTTGTTATATGATTCA CCCTGTTTTTCACAGCAAGG	Human enteric coronaviruse
PBHE_00006	CATGACGGCAGTTGCTTGTCAACCCCCGTACTGTTATTTTCGTAATTC TACTACCAACTATGTTGGTG	Human enteric coronaviruse
PBRh_00001	GGCTGAGTGATTACATCACAGGTTTGGGTAGAGCTTTTGGTGTCGGG TTCACTGACCAAATCTCAACAAA	Human rhinovirus
PBRh_00002	GAAAAGCTATTAGCTTGGTAGACAGAACTACCAACGTTAGGTATAGTG TGGATCAACTGGTCACGGCTAT	Human rhinovirus
PBRh_00003	GGCCAAGTAATAGCTAGACATAAGGTTAGGGAGTTTAACATAAATCCA GTCAACACGGCAACTAAGTCAA	Human rhinovirus
PBRh_00004	GATAACAAGGGCATGTTATTCACCAGTAATTTTGTTCTAGCCTCCACA AATTCTAACACACTAAGCCCCC	Human rhinovirus
PBRh_00005	GGCCAAGAAGTAAGGTTGTGTTTAGTACCACTCAGGGTTTACCAGTTA TGTTAACACCTGGATCTGGGCA	Human rhinovirus
PBRh_00006	GTAATGCGTAAGTGCGGGATGGGACCAACTACTTTGGGTGTCCGTGT TTCCTGTTTTTCTTTTGATTGCA	Human rhinovirus
PBRh_00007	TAAAAGAGGATTCAGAGCTGATGAGCGCCACTCTTTCCTTATACACCC TACCTTTCCTGTGGCTGAGATT	Human rhinovirus
PBRh_00008	GCAAGTTTCATCAGGGTTTATTAATAGTTGCCGCCATCCCAGAACATC AATTGGCATCTGCAACAAGTGG	Human rhinovirus
PBMP_00001	ATATATGAAGGAACACCAGTGGCGAAGGCGAAAACTTAGGCCATTAC TGACGCTTAGGCTTGAAAGTGTG	*Mycoplasma pneumoniae*
PBMP_00002	GCAGTAGGGAATTTTTCACAATGAGCGAAAGCTTGATGGAGCAATGC CGCGTGAACGATGAAGGTCTTTA	Mycoplasma pneumoniae
PBMP_00003	AACACATTAAGTATCTCGCCTGGGTAGTACATTCGCAAGAATGAAACT	Mycoplasma pneumoniae

	CAAACGGAATTGACGGGGACCC	
PBMP_00004	ACACCGTAAACGATAGATACTAGCTGTCGGGGCGATCCCCTCGGTAG TGAAGTTAACACATTAAGTATCT	Mycoplasma pneumoniae
PBMP_00005	ACATCCTTGGCAAAGTTATGGAAACATAATGGAGGTTAACCGAGTGAC AGGTGGTGCATGGTTGTCGTCA	Mycoplasma pneumoniae
PBR_00001	TTATAACTTAACCGTCGGCAGTTGGGTAAGAGACCACGTCCGATCAAT TGTCGAGGGCGCGTGGGAAGTG	Rubella virus
PBR_00002	ATACCCAGACCTGTGTTCACGCAGATGCAGGTCAGTGATCACCCAGC ACTCCACGCAATTTCGCGGTATA	Rubella virus
PBR_00003	AGAAACTCCTAGATGAGGTTCTTGCCCCCGGTGGGCCTTATAACTTAA CCGTCGGCAGTTGGGTAAGAGA	Rubella virus
PBR_00004	ATACCCAGACCTGTGTTCACGCAGATGCAGGTCAGTGATCACCCAGC ACTCCACGCAATTTCGCGGTATA	Rubella virus
PBR_00005	TCTTACTTCAACCCTGGCGGCAGCTACTACAAGCAGTACCACCCTAC CGCGTGCGAGGTTGAACCT	Rubella virus
PBM_00001	AAGGCTTGTTTCAGAGATTGCAATGCATACTACTGAGGACAGGATCAG TAGAGCAGTTGGACCCAGACAA	Measles virus
PBM_00002	AGGATCAGTAGAGCAGTTGGACCCAGACAAGCCCAAGTGTCATTCCT ACACGGTGATCAAAGTGAGAATG	Measles virus
PBM_00003	TCAGTAGAGCAGTTGGACCCAGACAAGCCCAAGTGTCATTCCTACAC GGTGATCAAAGTGAGAATG	Measles virus
PBM_00004	CCCAGGGAATGTACGGGGGAACTTACCTAGTTGAAAAGCCTAATCTG AGCAGCAAAGGATCAGAATTATC	Measles virus
PBM_00005	CCCAGGGAATGTACGGGGGAACTTACCTAGTTGAAAAGCCTAATCTG AGCAGCAAAGGATCAGAATTATC	Measles virus
PBRSV_00001	CAAACCCACAAACAAACCAACCACCAAAACCACAAACAAAAGAGACC CAAAAACACCAGCCAAAACGACG	Human respiratory syncytial virus
PBRSV_00002	GCAGCACTTGTAATAACCAAATTAGCAGCAGGAGACAGATCAGGTCTT ACAGCAGTAATTAGGAGGGCAA	Human respiratory syncytial virus
PBRSV_00003	CAAGAGGGGGTAGTAGAGTTGAAGGAATCTTTGCAGGATTGTTTATG	Human respiratory

	AATGCCTATGGTTCAGGGCAAGT	syncytial virus
PBRSV_00004	GACTTAACAGCAGAAGAATTGGAAGCCATAAAGAATCAACTCAACCCT AAAGAAGATGATGTAGAGCTTT	Human respiratory syncytial virus
PBRSV_00005	TCACAATCCACTGTGCTCGACACAACCACATTAGAACACACAATCCAA CAGCAATCCCTCCACTCAACCA	Human respiratory syncytial virus
PBRSV_00006	GACTTAACAGCAGAAGAATTGGAAGCCATAAAGAATCAACTCAACCCT AAAGAAGATGATGTAGAGCTTT	Human respiratory syncytial virus
PBPI_00001	GCCGACGACCATCAAGCGTAGCCAAACAAGATCAGAGAGAACACAGA ATTCAGAACTCCACAAATCAACA	Parainfluenza
PBPI_00002	CGACCCAAGATCATAGATCAAGTGAGGAGAGTGGAATCTCTAGGAGA ACAGGTGAGTCAAAAACTGAGAC	Parainfluenza
PBPI_00003	CGCAAATGAAGAGGGAACCAGCAACACATCAGTCGATGAGATGGCCA AGTTACTAGTAAGTCTTGGTGTA	Parainfluenza
PBPI_00004	CTCCTTGCAATGGCCATACGTAGTCCGGAATTATATCTCACTACAAAC GGTGTCAATGCTGATGTCAAGT	Parainfluenza
PBPI_00005	GAACAAAAACAGATGGGTTCATTGTCAAAACGAGAGACATGGAGTAT GAAAGAACCACAGAGTGGTTGTT	Parainfluenza
PBPI_00006	TGTTCCAAGGGCAAAGAGAGAATGCGGATCTAGAGGCATTGCTTCAG ACATATGGATATCCTGCATGTCT	Parainfluenza
PBPI_00007	GGTATATCCCTCTTCCCAGCCACATCATGACAAAAGGGGCATTTCTAG GTGGAGCAGATATCAAAGAATG	Parainfluenza
PBPI_00008	GTATAACAACCACATGTACATGCAACGGTATTGGCAATAGAATCAATC AACCACCTGATCAAGGAGTAAA	Parainfluenza
PBPI_00009	CCCAACCCATTCAAAACGAAAATCTCAAAAGAGATTGGCAACACAACA AACACTGAACATCATGCCAACC	Parainfluenza
PBME_00001	AAAAGTGTATCACAGAAGTTTGTTCATTGAGTATGGCAAAGCATTAGG CTCATCATCTACAGGCAGCAAA	Human metapneumovirus
PBME_00002	GAAAGTCTATTTGTTAATATATTCATGCAAGCTTATGGAGCCGGTCAA ACAATGCTAAGGTGGGGGGTCA	Human metapneumovirus
PBME_00003	ACGCTGTTGTGTGGAGAAATTCTGTATGCTAAACATGCTGATTACAAA	Human metapneumovirus

	TATGCTGCAGAAATAGGAATAC	
PBME_00004	TTAAGGAATCATCAGGTAATATCCCACAAAATCAGAGGCCCTCAGCAC CAGACACACCCATAATCTTATT	Human metapneumovirus
PBME_00005	TGAGCAATCAAAGGAGTGCAACATCAACATATCCACTACAAATTACCC ATGCAAAGTCAGCACAGGAAGA	Human metapneumovirus
PBME_00006	CTGTTCCATTGGCAGCAACAGAGTAGGGATCATCAAGCAGCTGAACA AAGGTTGCTCCTATATAACCAAC	Human metapneumovirus
PBME_00007	ACTTAATGACAGATGCTGAACTAGCCAGGGCCGTTTCTAACATGCCG ACATCTGCAGGACAAATAAAATT	Human metapneumovirus
PBME_00008	AAAAAAAGGGAAACTATGCTTGCCTCTTAAGAGAAGACCAAGGGTGG TATTGTCAGAATGCAGGGTCAAC	Human metapneumovirus
PBME_00009	GAAAAGAACACACCAGTTACAATACCAGCATTTATCAAATCGGTTTCT ATCAAAGAGAGTGAATCAGCCA	Human metapneumovirus
PBME_00010	CAAATCAGTTGGCAAAAAAACACATGATCTGATCGCATTATGTGATTTT ATGGATCTAGAAAAGAACACA	Human metapneumovirus
PBME_00011	CAGCTAAAGACACTGACTATAACTACTCTGTATGCTGCATCACAAAGT GGTCCAATACTAAAAGTGAATG	Human metapneumovirus
PBME_00012	AAAAGAACACACCAGTTACAATACCAGCATTTATCAAATCGGTTTCTAT CAAAGAGAGTGAATCAGCCAC	Human metapneumovirus
PBME_00013	CTATTATAGGAGAAAAAGTGAACACTGTATCTGAAACATTGGAATTAC CTACTATCAGTAGACCCACCAA	Human metapneumovirus
PBME_00014	AAGTTAGCATGGACAGACAAAGGTGGGGCAATCAAAACTGAAGCAAA GCAAACAATCAAAGTTATGGATC	Human metapneumovirus
PBME_00015	CAGGAAAATACACAAAGTTGGAGAAAGATGCTCTAGACTTGCTTTCAG ACAATGAAGAAGAAGATGCAGA	Human metapneumovirus
PBME_00016	CTAATAGCAGACATAATAAAAGAAGCCAAGGGAAAAGCAGCAGAAAT GATGGAAGAAGAAATGAACCAGC	Human metapneumovirus
PBCP_00001	ACCCTTATCGTTAGTTGCCAGCACTTAGGGTGGGAACTCTAACGAGA CTGCCTGGGTTAACCAGGAGGAA	Chlamydophila pneumoniae
PBCP_00002	ATAAGAGAGGTTGGCTAATATCCAATTGATTTGAGCGTACCAGGTAAA	Chlamydophila

	GAAGCACCGGCTAACTCCGTGC	pneumoniae
PBCP_00003	CATGGGATCTTAAGTTTTAGTTGAATACTTCTGGAAAGTTGAACGATA CAGGGTGATAGTCCCGTAAACG	Chlamydophila pneumoniae
PBCP_00004	GGGTGCTAGCGTTAATCGGATTTATTGGGCGTAAAGGGCGTGTAGGC GGAAAGGAAAGTTAGATGTTAAA	Chlamydophila pneumoniae
PBCP_00005	GCCAGGGAGTTAAGTTAAACGGCGAGATTAAGGGATTTACATTCCGG AGTCGAAGCGAAAGCGAGTTTTA	Chlamydophila pneumoniae
PBCP_00006	GCCAGGGAGTTAAGTTAAACGGCGAGATTAAGGGATTTACATTCCGG AGTCGAAGCGAAAGCGAGTTTTA	Chlamydophila pneumoniae

In some embodiments, the non-SARS-CoV infectious organism is an infectious organism damaging an infectious host's immune system. Such organism includes, but not limited to, a hepatitis virus, a transfusion transmitting virus (TTV), a human immunodeficiency virus (HIV), a parvovirus, a human cytomegalovirus (HCMV), an Epstein-Barr virus (EBV) and a tre-ponema palidum. The hepatitis virus can be hepatitis virus A (HAV), hepatitis virus B (HBV), hepatitis virus C (HCV), hepatitis virus D (HDV), hepatitis virus E (HEV), or hepatitis virus G (HGV). The HIV can be HIV I. The parvovirus can be parvovirus B19. Exemplary probes are set forth in Table 16.

Table 16. Exemplary probes for non-SARS-CoV infectious organisms damaging host's immune system

Id	sequence(5'-3')	species
PBHAV_00001	GGTGTTGAACCTGAGAAAAATATTTACACCAAACCTGTGGCCTCAGATT ATTGGGATGGATATAGTGGAC	HAV
PBHAV_00002	ACTGAGGAGCATGAAATAATGAAGTTTTCTTGGAGAGGAGTGACTGCTG ATACTAGGGCTTTGAGAAGAT	HAV
PBHAV_00003	CATGGCGTGACTAAGCCCAAACAAGTGATTAAATTGGATGCAGATCCAG TAGAGTCCCAGTCAACTCTAG	HAV
PBHAV_00004	GTGCAGTGATGGACATTACAGGAGTGCAGTCAACCTTGAGATTTCGTGT	HAV

	TCCTTGGATTTCTGATACACC	
PBHAV_00005	CCAAAAGAGATTTAATTTGGTTGGATGAAAATGGTTTGCTGTTAGGAGTT CACCCAAGATTGGCCCAGAG	HAV
PBHAV_00006	AGAGATGCTTTGGATAGGGTAACAGCGGCGGATATTGGTGAGTTGTTAA GACAAAAACCATTCAACGCCG	HAV
PBHBV_00001	GCTGGATGTGTCTGCGGCGTTTTATCATATTCCTCTTCATCCTGCTGCTA TGCCTCATCTTCTTATTGGT	HBV
PBHBV_00002	ATATACATCCTTTCCATAGCTGCTAGGTTGTACTGCCAACTAGATTCTTC GCGGGACGTCCTTTGTCTAC	HBV
PBHBV_00003	ATTCTTTCCCGATCATCAGTTGGACCCTGCATTCGGAGCCAATTCAAAC AATCCAGATTGGGACTTCAAC	HBV
PBHBV_00004	CTCATGTTGCTGTACAAAACCTACGGATGGAAATTGCACCTGTATTCCC ATCCCATCATCTTGGGCTTTC	HBV
PBHBV_00005	AGAGTCTAGACTCGTGGTGGACTTCTCTCAATTTTCTAGGGGGAGCACC CGTGTGTCTTGGCCAAAATTC	HBV
PBHBV_00006	CCTTGGATGGCTTTGGGGCATGGACATTGACCCTTATAAAGAATTTGGA GCTACTGTGGAGTTACTCTCA	HBV
PBHCV_00001	TGGGAGACAGCAAGACACACTCCAGTCAATTCCTGGCTAGGCAACATA ATCATGTTTGCCCCCACACTGT	HCV
PBHCV_00002	TGAGCGACTTTAAGACCTGGCTGAAAGCCAAGCTCATGCCACAACTGC CTGGGATTCCCTTTGTGT	HCV
PBHCV_00003	TATAGATGCCCACTTTCTATCCCAGACAAAGCAGAGTGGGGAGAACTTT CCTTACCTGGTAGCGTACCAA	HCV
PBHCV_00004	TAACAACACCAGGCCACCGCTGGGCAATTGGTTCGGTTGTACCTGGAT GAACTCAACTGGATTCACCAAA	HCV
PBHCV_00005	TTTATCCCTGTGGAGAACCTAGAGACAACCATGAGATCCCCGGTGTTCA CGGACAACTCCTCTCCACCAG	HCV
PBHCV_00006	TTTATCCCTGTGGAGAACCTAGAGACAACCATGAGATCCCCGGTGTTCA CGGACAACTCCTCTCCACCAG	HCV
PBHDV_00001	TTCCCTTCTCTCGTCTTCCTCGGTCAACCTCTTAAGTTCCTCTTCTTCTT	HDV

	CCTTGCTGAGGTGCTTCCCT	
PBHDV_00002	TAAGCCCATAGCGATAGGGAGAGATGCTAGGAGTTAGAGGAGACCGAA GCGAGGAGGAAAGCAAAGAGAG	HDV
PBHDV_00003	TTGGAGAGCACTCCGGCCGAAAGGTCGAGGTACCCAGAAGGAGGAAT CTCACGGAGAAAAGCAGACAAAT	HDV
PBHDV_00004	TTAAGTTCCTCTTCTTCTTCCTTGCTGAGGTGCTTCCCTCCCGCGGCCA GCTGCTTTCTCTTGTTCTCGA	HDV
PBHDV_00005	AAAAAGAGAAAGCAAGAGACGGACGATTTCCCCATGACTCTGGAGACA TCCTGGAAGGGGAAAGAAGGAA	HDV
PBHDV_00006	AAGTTCCTCTTCTTCTTCCTTGCTGAGGTGCTTCCCTCCCGCGGCCAGC TGCTTTCTCTTGTTCTCGAGG	HDV
PBHGV_00001	TCATATCATGCATCATTGGACACGGCCCCCTTCTGCTCCACTTGGCTTG CTGAGTGCAATGCAGAT	HGV
PBHGV_00002	TAAAGTGGGAAAGTGAGTTTTGGAGATGGACTGAACAGCTGGCCTCCA ACTACTGGATTCTGGAATACCT	HGV
PBHGV_00003	TAGGTCGTAAATCCCGGTCACCTTGGTAGCCACTATAGGTGGGTCTTAA GAGAAGGTTAAGATTCCTCTT	HGV
PBHGV_00004	TTCTTGGTTTGCCTCCACCAGTGGTCGCGACTCGAAGATAGATGTGTGG AGTTTAGTGCCAGTTGG	HGV
PBHGV_00005	TCCAACTACTGGATTCTGGAATACCTCTGGAAGGTCCCATTTGATTTCT GGAGAGGCGTGATAAGCCTGA	HGV
PBHGV_00006	ACGTTACCAAGGTCTTCATGTATCCCGGACAGTTACTTTCAGCAAGTTG ACTATTGCGACAAGGTCTCAG	HGV
PBTTV_00001	TGTCAGTAACAGGGGTCGCCATAGACTTCGGCCTCCATTTTACCTTGTA AAAACTACCAAAATGGCCGTT	TTV
PBTTV_00002	ATGTCATCCATTTCCTGGGCCGGGTCTACGTCCTCATATAAGTAACTGC ACTTCCGAATGGCTGAGTTTT	TTV
PBTTV_00003	GGGATCTAGCATCCTTATTTCAAATAGCACCATAAACATGTTTGGTGACC CCAAACCTTACAACCCTTCC	TTV
PBTTV_00004	TGTTAGAAATCCCTGCAAAGAAACCCACTCCTCGGGCAATAGAGTCCCT	TTV

	AGAAGCTTACAAATCGTTGAC	
PBTTV_00005	TCAAGGATTGACGTAAAGGTTAAAGGTCATCCTCGGCGGAAGCTACACA AAATGGTGGACAACATCTTCC	TTV
PBB19_00001	GGCATGGTTAACTGGAATAATGAAAACTTTCCATTTAATGATGTAGCAG GGAAAAGCTTGGTGGTCTGGG	B19
PBB19_00002	GGCAAGAAAAATACACTGTGGTTTTATGGGCCGCCAAGTACAGGAAAAA CAAACTTGGCAATGGCCATTG	B19
PBB19_00003	GCCATTTCTCATGGTCAGACCACTTATGGTAACGCTGAAGACAAAGAGT ATCAGCAAGGAGTGGGTAGAT	B19
PBB19_00004	AATTTCGAGAATTTACCCCAGATTTGGTGCGGTGTAGCTGCCATGTGGG AGCTTCTAATCCCTTTTCTGT	B19
PBHCMV_00001	AGGTGCGCAACGCTTTTATGAAGGTAAAGCCCGTGGCCCAGGAGATTA TCCGTATCTGCATACTCGCTAA	HCMV
PBHCMV_00002	TAAACGACATGTATCTGTTGTTGACGCTGCGACACTTGCAGCTGCGACA CGCGCTGGAGCTACAAATGAT	HCMV
PBHCMV_00003	CAAAGCAGCGTCAACAACAGCCACACAGAAACCTACGTGGAGACGACA CGGGACTTTTTATTGACGGAGA	HCMV
PBHCMV_00004	TGCTCCAAAGCAGCGTCAACAACAGCCACACAGAAACCTACGTGGAGA CGACACGGGACTTTTTATTGAC	HCMV
PBEBV_00001	GAGTTAAAAGCAACTACTGTTTATTTTCCAAAATGAGCTGGGTATAGTTG ATGATCTGTAGGCGCAGCTC	EBV
PBEBV_00002	ACAGTGACAGTGGGAGAAACACGGCCTCTGAGACATGTATGGGGGTGT TCATCTCACGCAGAAAATCTTT	EBV
PBEBV_00003	TGAAGAAGTCCCGTAGTGAAAAATGGGATCTGTCTACACCATGTCTGGT GTGCCGGGAACATATTGATCG	EBV
PBEBV_00004	TGAAGAAGTCCCGTAGTGAAAAATGGGATCTGTCTACACCATGTCTGGT GTGCCGGGAACATATTGATCG	EBV
PBHIV1_00001	ATTATTGTCTGGTATAGTGCAGCAGCAGAACAATTTGCTGAGGGCTATT GAGGCGCAACAGCATCTGTTG	HIV1
PBHIV1_00002	GCAACCCTCTATTGTGTGCATCAAAGGATAGAGATAAAAGACACCAAGG	HIV1

	AAGCTTTAGACAAGATAGAGG	
PBHIV1_00003	TGTATGTAGGATCTGACTTAGAAATAGGGCAGCATAGAACAAAAATAGA GGAGCTGAGACAACATCTGTT	HIV1
PBHIV1_00004	GGAATGCTAGTTGGAGTAATAAATCTCTGGAACAGATTTGGAATCACAC GACCTGGATGGAGTGGGACAG	HIV1
PBTP_00001	TACCTTGAAAGACGTTACCGCCAAAATGCTCATCAAAAGAACGAGGACC ATGCTGACAGCACCCGCGACA	TP
PBTP_00002	TTTCGTGATCCTTTTCCTTTTCCTGTAGCTCAGCGTCCTTTTTATCTAATT CCTCTGCACGCTCCCCGAG	TP
PBTP_00003	TCTTTCTGACTCGCGCAAAAGGCATTACTGGAACACTATTTTAGCCATGT GGTGGCTCCCTGCTATCTTA	TP
PBTP_00004	ACCTTGAAAGACGTTACCGCCAAAATGCTCATCAAAAGAACGAGGACCA TGCTGACAGCACCCGCGACAA	TP
PBHEV_00001	AATAATTCACGCCGTCGCTCCTGATTATAGGTTGGAACATAACCCAAAG ATGCTTGAGGCTGCCTACCGG	HEV
PBHEV_00002	TTTGTTGACGGGGCGGTTTTAGAGACTAATGGCCCAGAGCGCCACAAT CTCTCTTTTGATGCCAGTCAGA	HEV
PBHEV_00003	ATTTTACTAGTACTAATGGTGTCGGTGAGATCGGCCGCGGGATAGCGC TTACCCTGTTTAACCTTGCTGA	HEV
PBHEV_00004	AGTCCACTTACGGCTCTTCGACCGGCCCAGTCTATGTCTCTGACTCTGT GACCTTGGTTAATGTAG	HEV

In some embodiments, the non-SARS-CoV infectious organism is a non-SARS-CoV coronaviridae virus. Such virus includes, but not limited to, an avian infectious bronchitis virus, an avian infectious laryngotracheitis virus, a murine hepatitis virus, an equine coronaviruse, a canine coronaviruse, a feline coronaviruse, a porcine epidemic diarrhea virus, a porcine transmissible gastroenteritis virus, a bovine coronaviruse, a feline infectious peritonitis virus, a rat coronaviruse, a neonatal calf diarrhea coronaviruse, a porcine hemagglutinating encephalomyelitis virus, a puffinosis

virus, a turkey coronaviruse and a sialodacryoadenitis virus of rat. Exemplary probes for these viruses are set forth in Table 17.

Table 17. Exemplary probes for non-SARS-CoV coronaviridae virus

seqid	sequence(5'-3')
PBIBV_00001	GGTATAGTGTGGGTTGCTGCTAAGGGTGCTGATACTAAATCTAGATCCAATCAGGGTACAAGAGATCCTG
PBIBV_00002	GGTATAGTGTGGGTTGCTGCTAAGGGTGCTGATACTAAATCTAGATCCAATCAGGGTACAAGAGATCCTG
PBMHV_00001	CCAGCCCAAGCAAGTAACGAAGCAAAGTGCCAAAGAAGTCAGGCAGAAAATTTTAAACAAGCCTCGCCAA
PBMHV_00002	TCTAAACTTTAAGGATGTCTTTTGTTCCTGGGCAAGAAAATGCCGGTGGCAGAAGCTCCTCTGTAAACCG
PBEQ_00001	AGGATCAAGAAATAGATCCAATTCCGGCACTAGAACACCCACCTCTGGTGTGACATCTGATATGGCTGAT
PBEQ_00002	TTTAAAACAGCCGATGGCAATCAACGCCAATTGTTGCCACGCTGGTATTTTTACTACTTGGGAACAGGCC
PBCA_00001	TTGGAACTTATGTCCGAGAGACTTTGTACCCAAAGGAATAGGTAACAAGGATCAACAGATTGGTTATTGG
PBCA_00002	GCTGAATGTGTTCCATCTGTATCTAGCATTCTGTTTGGAAGCTATTGGACTGCAAAGGAAGATGGCGACC
PBFE_00001	CACCACCCTCGAACAAGGAGCTAAATTTTGGTATGTATGTCCGAGAGACTTTGTTCCCAAGGGAATAGGT
PBFE_00002	GGCACTCGTGGAACCAACAATGAATCCGAACCATTGAGATTTGATGGTAAGATACCACCACAATTCCAGC
PBPEDV_00001	CTGATCCAAATGTTGAGCTTCTTGTTGCACAGGTGGATGCATTTAAAACTGGGAATGCAAAACCCCAGAG
PBPEDV_00002	ATGAGCAAATTCGCTGGCGTATGCGCCGTGGTGAGCGAATTGAACAACCTTCAAATTGGCATTTCTACTA
PBPTGV_00001	GAGAGACTTTGTACCCAAAGGAATAGGTAACAGGGATCAACAGATTGGTTATTGGAATAGACAAACTCGC
PBPTGV_00002	GATGGTGACCAGATAGAAGTCACGTTCACACACAAATACCACTTGCCAAAGGATGATCCTAAAACTGGAC
PBBOV_00001	TATTTTTACTATCTTGGAACAGGACCGCATGCCAAAGACCAGTATGGCACCGACATTGACGGAGTCTACT
PBBOV_00002	AGAACCCCTACCTCTGGTGTAACACCTGATATGGCTGATCAAATTGCTAGTCTTGTTCTGGCTAAACTTG
PBFIPV_00001	GAGTGTGGTTAATCAACAGGGTGAAGCGCTGAGTCAACTTACCAGTCAGTTACAGAAAAACTTCCAGGCT
PBFIPV_00002	CCGGCATTGTAGATGGTAATAAGATGGCCATGTACACAGCATCTTTAATTGGAGGTATGGCTTTGGGCTC
PBR_00001	AAATGTTAAAACTTGGAACTAGTGATCCACAGTTCCCCATTCTTGCAGAGTTGGCCCCAACACCTGGTGC
PBR_00002	CCCATTACTCTTGGTTTTCGGGCATTACCCAATTTCAAAAGGGAAAGGAGTTCCAGTTTGCAGATGGGCA
PBPHEV_00001	TAGTAACCAGGCTGATATTAATACCCCGGCTGACATTGTCGATCGGGATCCAAGTAGCGATGAGGCTATT
PBPHEV_00002	TTCTTTTAAAACAGCCGATGGCAATCAGCGTCAACTGCTGCCACGATGGTACTTTTACTACCTGGGAACA
PBPV_00001	GTGGTTCCCCATTACTCCTGGTTTTCTGGCATTACCCAATTCCAGAAGGGAAAGGAGTTTAAGTTTGCAG
PBPV_00002	AAGAAGTCAGGCAGAAAATTTTAAACAAGCCTCGCCAAAAGAGGACTCCAAACAAGCAGTGCCCAGTGCA

PBTK_00001	TTTGGTGATGACAAGATGAATGAGGAAGGTATTAAGGATGGGCGTGTTACGGCAATGCTCAACCTAGTCC
PBTK_00002	TTTGGTGATGACAAGATGAATGAGGAAGGTATTAAGGATGGGCGTGTTACGGCAATGCTCAACCTAGTCC
PBSDAV_00001	AGCCTGCCTCTACTGTAAAACCTGATATGGCCGAAGAAATTGCTGCTCTTGTTTTGGCTAAGCTAGGCAA
PBSDAV_00002	CCCCATTCTTGCAGAGTTGGCCCCAACACCTGGTGCCTTCTTCTTTGGATCTAAATTAGAATTGGTCAAA

The oligonucleotide probes and the target SARS-CoV and any non-SARS-CoV infectious organism nucleotide sequences can be any suitable length. Preferably, the oligonucleotide probes and the target SARS-CoV and any non-SARS-CoV infectious organism nucleotide sequences have a length of at least 7, 10, 20, 30, 40, 50, 60, 80, 90, 100 or more than 100 nucleotides.

The oligonucleotide probes and primers can be prepared by any suitable methods, e.g., chemical synthesis, recombinant methods and/or both (*See generally*, Ausubel et al., (Ed.), Current Protocols in Molecular Biology, John Wiley & Sons, Inc. (2000)).

Any suitable support can be used in the present chips. For example, the support can comprise a surface that is selected from the group consisting of a silicon, a plastic, a glass, a ceramic, a rubber, and a polymer surface.

C. Methods for assaying for a SARS-CoV and a non-SARS-CoV infectious organism

In another aspect, the present invention is directed to a method for assaying for a SARS-CoV and a non-SARS-CoV infectious organism in a sample, which methods comprises: a) providing an above-described chip; b) contacting said chip with a sample containing or suspected of containing a nucleotide sequence of a SARS-CoV and a non-SARS-CoV infectious organism under conditions suitable for nucleic acid hybridization; and c) assessing hybrids formed between said nucleotide sequence of said SARS-CoV or said non-SARS-CoV infectious organism, if present in said sample, and said oligonucleotide probe complementary to a nucleotide sequence of said SARS-CoV genome or said oligonucleotide probe complementary to a nucleotide sequence of said non-SARS-CoV infectious organism genome, whereby detection of one or both of said

hybrids indicates the presence of said SARS-CoV and/or said non-SARS-CoV infectious organism in said sample.

In some embodiments, the SARS-CoV is assayed by: a) providing a chip comprising a support suitable for use in nucleic acid hybridization having immobilized thereon at least two oligonucleotide probes complementary to at least two different nucleotide sequences of SARS-CoV genome, each of said two different nucleotide sequences comprising at least 10 nucleotide; b) contacting said chip with a sample containing or suspected of containing a SARS-CoV nucleotide sequence under conditions suitable for nucleic acid hybridization; and c) assessing hybrids formed between said SARS-CoV nucleotide sequence, if present in said sample, and said at least two oligonucleotide probes complementary to two different nucleotide sequences of SARS-CoV genome, respectively, to determine the presence, absence or amount of said SARS-CoV in said sample, whereby detection of one or both said hybrids indicates the presence of said SARS-CoV in said sample.

In a specific embodiment, the present methods comprise: a) providing a chip comprising a nucleotide sequence of at least 10 nucleotides located within a conserved region of SARS-CoV genome and a nucleotide sequence of at least 10 nucleotides located within a variable region of SARS-CoV genome, or a nucleotide sequence of at least 10 nucleotides located within a structural protein coding gene of SARS-CoV genome and a nucleotide sequence of at least 10 nucleotides located within a non-structural protein coding gene of SARS-CoV genome; b) contacting said chip with a sample containing or suspected of containing a SARS-CoV nucleotide sequence under conditions suitable for nucleic acid hybridization; and c) assessing hybrids formed between said SARS-CoV nucleotide sequence, if present in said sample, and i) said oligonucleotide probe complementary to a nucleotide sequence located within a conserved region of SARS-CoV genome and an oligonucleotide probe complementary to a nucleotide sequence located within a variable region of SARS-CoV genome, respectively; or ii) said oligonucleotide probe complementary to a nucleotide sequence located within a structural protein coding gene of SARS-CoV genome and an oligonucleotide probe complementary to a nucleotide sequence located within a

non-structural protein coding gene of SARS-CoV genome, to determine the presence, absence or amount of said SARS-CoV in said sample, whereby detection of one or both said hybrids indicates the presence of said SARS-CoV in said sample.

In another specific embodiment, the present methods comprise: a) providing a chip comprising an oligonucleotide probe complementary to a nucleotide sequence within a conserved region of SARS-CoV genome, an oligonucleotide probe, complementary to a nucleotide sequence located within a variable region of SARS-CoV genome, at least one of the following three oligonucleotide probes: an immobilization control probe that is labeled and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip, a positive control probe that is not complementary to any SARS-CoV sequence but is complementary to a non-SARS-CoV-sequence contained in the sample and a negative control probe that is not complementary to any nucleotide sequence contained in the sample, and a blank spot; b) contacting said chip with a sample containing or suspected of containing a SARS-CoV nucleotide sequence under conditions suitable for nucleic acid hybridization; and c) assessing: (i) hybrids formed between said SARS-CoV nucleotide sequence, if present in the sample, and the oligonucleotide probe complementary to a nucleotide sequence within a conserved region of SARS-CoV genome and an oligonucleotide probe complementary to a nucleotide sequence located within a variable region of SARS-CoV genome, respectively; (ii) a label comprised in the immobilization control probe, or a hybrid(s) involving the positive control probe and/or the negative control probe; and (iii) a signal at said blank spot to determine the presence, absence or amount of said SARS-CoV in a sample.

Preferably, the present chips comprise two oligonucleotide probes complementary to two different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV, an oligonucleotide probe complementary to a nucleotide sequence located within the N gene of SARS-CoV, an oligonucleotide probe complementary to a nucleotide sequence located within the S gene of SARS-CoV, an immobilization control probe, a positive control probe and a negative control probe and the presence of the SARS-CoV is determined when: a) a positive hybridization signal is

detected using at least one of the two different nucleotide sequences located within the Replicase 1 A or 1B gene of SARS-CoV, the oligonucleotide probe complementary to a nucleotide sequence located within the N gene of SARS-CoV and the oligonucleotide probe complementary to a nucleotide sequence located within the S gene of SARS-CoV; b) a positive signal is detected from the immobilization control probe; c) a positive hybridization signal is detected using the positive control probe; d) a positive hybridization signal is not detected using the negative control probe; and e) a positive hybridization signal is not detected at the blank spot.

The inclusion of a target sequence in a variable region of SARS-CoV enables an assessment of possible mutation of the SARS-CoV. For example, detecting a positive hybridization signal using at least one of the two different nucleotide sequences located within the Replicase 1A or 1B gene of SARS-CoV, or the oligonucleotide probe complementary to a nucleotide sequence located within the N gene of SARS-CoV, while not detecting a positive hybridization signal using the oligonucleotide probe complementary to a nucleotide sequence located within the S gene of SARS-CoV indicates a mutation(s) of the SARS-CoV.

The present methods can be used for any suitable prognosis and diagnosis purpose. In one example, the present method is used to positively identify SARS-CoV infected patients from a population of patients who have SARS-like symptoms, *e.g.*, fever or elevated temperature, nonproductive cough, myalgia, dyspnea, elevated lactate dehydrogenase, hypocalcemia, and lymphopenia (Booth et al., *JAMA*, 2003 May 6; [epub ahead of print]). The present chips, methods and kits can further comprise assaying for elevated lactate dehydrogenase, hypocalcemia, and lymphopenia, etc.

In another example, a chip further comprising an oligonucleotide probe complementary to a nucleotide sequence of a coronaviruse not related to the SARS-CoV is used and the method is used to positively identify SARS-CoV infected patients from patients who have been infected with a coronaviruse not related to the SARS, *e.g.*, a coronaviruse that infects an avian species, *e.g.*, Avian infectious bronchitis virus and Avian infectious laryngotracheitis virus, an equine species, *e.g.*, Equine coronaviruse, a canine species, *e.g.*, Canine coronaviruse, a feline species, *e.g.*, Feline coronaviruse and

Feline infectious peritonitis virus, a porcine species, *e.g.*, Porcine epidemic diarrhea virus, Porcine transmissible gastroenteritis virus and Porcine hemagglutinating encephalomyelitis virus, a calf species, *e.g.*, Neonatal calf diarrhea coronaviruse, a bovine species, *e.g.*, Bovine coronaviruse, a murine species, *e.g.*, Murine hepatitis virus, a puffinosis species, *e.g.*, Puffinosis virus, a rat species, *e.g.*, Rat coronaviruse and a Sialodacryoadenitis virus of rat, *e.g.*, a turkey species *e.g.*, Turkey coronaviruse, or a human species, *e.g.*, Human enteric coronaviruse.

In still another example, a chip comprising an oligonucleotide probes complementary to a highly expressed nucleotide sequence of SARS-CoV genome is used and the method is used to diagnose early-stage SARS patients, *e.g.*, SARS patients who have been infected with SARS-CoV from about less than one day to about three days.

In yet another example, the present methods are used to monitor treatment of SARS, *e.g.*, treatment with an interferon or an agent that inhibits the replication of a variety of RNA viruses such as ribavirin. The present methods can also be used to assess potential anti-SARS-CoV agent in a drug screening assay.

The method of the invention can be used to determine whether a subject is infected by a SARS-CoV and/or a non-SARS-CoV infectious organism causing SARS-like symptoms. Non-SARS-CoV infectious organism that causing SARS-like symptoms includes, but not limited to, a human coronaviruse 229E, a human coronaviruse OC43, a human enteric coronaviruse, an influenza virus, a parainfluenza virus, a respiratory sncytical virus, a human metapneumovirus, a rhinovirus, an adenoviruse, a mycoplasma pneumoniae, a chlamydia pneumoniae, a measles virus and a rubella virus. The influenza virus can be influenza virus A or influenza virus B. The parainfluenza virus can be parainfluenza virus 1, parainfluenza virus 2, parainfluenza virus 3 or parainfluenza virus 4.

The method of the invention can also be used to determine whether a subject is infected by a SARS-CoV and/or a non-SARS-CoV infectious organism damaging the subject's immune system. The non-SARS-CoV infectious organism damaging subject's immune system includes, but not limited to, a hepatitis virus, a transfusion transmitting virus (TTV), a human immunodeficiency virus (HIV), a parvovirus, a human

cytomegalovirus (HCMV), an Epstein-Barr virus (EBV) and a tre-ponema palidum. The hepatitis virus can be hepatitis virus A (HAV), hepatitis virus B (HBV), hepatitis virus C (HCV), hepatitis virus D (HDV), hepatitis virus E (HEV), or hepatitis virus G (HGV). The HIV can be HIV I. The parvovirus can be parvovirus B19.

The method of the invention can also be used to determine whether a subject is infected by a SARS-CoV and/or a non-SARS-CoV coronaviridae virus. The non-SARS-CoV coronaviridae virus includes, but not limited to, an avian infectious bronchitis virus, an avian infectious laryngotracheitis virus, a murine hepatitis virus, an equine coronaviruse, a canine coronaviruse, a feline coronaviruse, a porcine epidemic diarrhea virus, a porcine transmissible gastroenteritis virus, a bovine coronaviruse, a feline infectious peritonitis virus, a rat coronaviruse, a neonatal calf diarrhea coronaviruse, a porcine hemagglutinating encephalomyelitis virus, a puffinosis virus, a turkey coronaviruse and a sialodacryoadenitis virus of rat.

Any suitable SARS-CoV or non-SARS-CoV infectious organism nucleotide sequence can be assayed. For example, the SARS-CoV or the non-SARS-CoV infectious organism nucleotide sequence to be assayed can be a SARS-CoV RNA or a non-SARS-CoV infectious organism genomic sequence or a DNA sequence amplified from an extracted SARS-CoV RNA or a non-SARS-CoV infectious organism genomic sequence.

The SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence can be prepared by any suitable methods. For example, the SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence can be extracted from a SARS-CoV or the non-SARS-CoV infectious organism infected cell or other materials using the QIAamp Viral RNA kit, the Chomczynski-Sacchi technique or TRIzol (De Paula et al., *J. Virol. Methods*, 98(2):119-25 (2001)). Preferably, the SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence is extracted from a SARS-CoV or the non-SARS-CoV infectious organism infected cell or other materials using the QIAamp Viral RNA kit. The SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence can be extracted from any suitable source. For example, the SARS-CoV RNA or the non-SARS-CoV infectious organism genomic

sequence can be extracted from a sputum or saliva sample. In another example, the SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence can be extracted from a lymphocyte of a blood sample.

The SARS-CoV RNA or the non-SARS-CoV infectious organism genomic sequence can be amplified by any suitable methods, *e.g.*, PCR. Preferably, a label is incorporated into the amplified DNA sequence during the PCR. Any suitable PCR can be used, *e.g.*, conventional, multiplex, nested PCR or RT-PCR. In one example, the PCR can comprise a two-step nested PCR, the first step being a RT-PCR and the second step being a conventional PCR. In another example, the PCR can comprise a one-step, multiplex RT-PCR using a plurality of 5' and 3' specific primers, each of the specific primers comprising a specific sequence complementary to its target sequence to be amplified and a common sequence, and a 5' and a 3' universal primer, the 5' universal primer being complementary to the common sequence of the 5' specific primers and the 3' universal primer being complementary to the common sequence of the 3' specific primers, and wherein in the PCR, the concentration of the 5' and 3' universal primers equals to or is higher than the concentration of the 5' and 3' specific primers, respectively. Preferably, the 3' universal primer and/or the 5' universal primer is labeled, *e.g.*, a fluorescent label. In still another example, the PCR comprises a multiple step nested PCR or RT-PCR. In yet another example, the PCR is conducted using at least one of the following pairs of primers for SARS-CoV set forth in Table 18.

Table 18. Exemplary SARS-CoV primers

Id	sequence(5'-3')	region
PMSL_00005	CACGTCTCCCAAATGCTTGAGTGACG	SARS-Cov Nucleocapsid gene
PMSU_00006	CCTCGAGGCCAGGGCGTTCC	SARS-Cov Nucleocapsid gene
PMV_00039	TCACTTGCTTCCGTTGAGGTCGGGGACCAAGACCTAATCAGA	SARS-Cov Nucleocapsid gene
PMV_00040	GGTTTCGGATGTTACAGCGTAGCCGCAGGAAGAAGAGTCACAG	SARS-Cov Nucleocapsid gene
PMV_00041	TCACTTGCTTCCGTTGAGGAGGCCAGGGCGTTCCAATC	SARS-Cov Nucleocapsid gene
PMV_00042	GGTTTCGGATGTTACAGCGTCAATAGCGCGAGGGCAGTTTC	SARS-Cov Nucleocapsid gene
PMV_00043	TCACTTGCTTCCGTTGAGGGGCACCCGCAATCCTAATAACAA	SARS-Cov Nucleocapsid gene

id	sequence(5'-3')	region
PMV_00044	GGTTTCGGATGTTACAGCGTAGCCGCAGGAAGAAGAGTCACAG	SARS-Cov Nucleocapsid gene
PMV_00090	TCGGGGACCAAGACCTAATCAGA	SARS-Cov Nucleocapsid gene
PMV_00091	AGCCGCAGGAAGAAGAGTCACAG	SARS-Cov Nucleocapsid gene
PMV_00092	AGGCCAGGGCGTTCCAATC	SARS-Cov Nucleocapsid gene
PMV_00093	CAATAGCGCGAGGGCAGTTTC	SARS-Cov Nucleocapsid gene
PMV_00094	GGCACCCGCAATCCTAATAACAA	SARS-Cov Nucleocapsid gene
PMV_00095	AGCCGCAGGAAGAAGAGTCACAG	SARS-Cov Nucleocapsid gene
PMSL_00001	ACATCACAGCTTCTACACCCGTTAAGGT	SARS-Cov Replicase 1A
PMSL_00002	ATACAGAATACATAGATTGCTGTTATCC	SARS-Cov Replicase 1A
PMSU_00002*	GCATCGTTGACTATGGTGTCCGATTCT	SARS-Cov Replicase 1A
PMSU_00003	GCTGCATTGGTTTGTTATATCGTTATGC	SARS-Cov Replicase 1A
PMV_00023	TCACTTGCTTCCGTTGAGGAGCCGCTTGTCACAATGCCAATT	SARS-Cov Replicase 1A
PMV_00024	GGTTTCGGATGTTACAGCGTCATCACCAAGCTCGCCAACAGTT	SARS-Cov Replicase 1A
PMV_00025	TCACTTGCTTCCGTTGAGGAGGTTGCCATCATTTTGGCATCTT	SARS-Cov Replicase 1A
PMV_00026	GGTTTCGGATGTTACAGCGTCTTTGCGCCAGCGATAGTGACTT	SARS-Cov Replicase 1A
PMV_00027	TCACTTGCTTCCGTTGAGGATGGCACCCGTTTCTGCAATGG	SARS-Cov Replicase 1A
PMV_00028	GGTTTCGGATGTTACAGCGTTCGGGCAGCTGACACGAATGTAGA	SARS-Cov Replicase 1A
PMV_00029	TCACTTGCTTCCGTTGAGGGAATGGCGATGTAGTGGCTATTGA	SARS-Cov Replicase 1A
PMV_00030	GGTTTCGGATGTTACAGCGTTAATGCCGGCATCCAAACATAAT	SARS-Cov Replicase 1A
PMV_00031	TCACTTGCTTCCGTTGAGGTAGCCAGCGTGGTGGTTCATACAA	SARS-Cov Replicase 1A
PMV_00032	GGTTTCGGATGTTACAGCGTCTCCCGGCAGAAAGCTGTAAGCT	SARS-Cov Replicase 1A
PMV_00033	TCACTTGCTTCCGTTGAGGTATAGAGCCCGTGCTGGTGATGC	SARS-Cov Replicase 1A
PMV_00034	GGTTTCGGATGTTACAGCGTATCGCCATTCAAGTCTGGGAAGAA	SARS-Cov Replicase 1A
PMV_00035	TCACTTGCTTCCGTTGAGGTGGCTCAGGCCATACTGGCATTAC	SARS-Cov Replicase 1A
PMV_00036	GGTTTCGGATGTTACAGCGTTTTGCGCCAGCGATAGTGACTTG	SARS-Cov Replicase 1A
PMV_00037	TCACTTGCTTCCGTTGAGGTTCCCGTCAGGCAAAGTTGAAGG	SARS-Cov Replicase 1A
PMV_00038	GGTTTCGGATGTTACAGCGTGACGGCAATTCCTGTTTGAGCAGA	SARS-Cov Replicase 1A
PMV_00074	AGCCGCTTGTCACAATGCCAATT	SARS-Cov Replicase 1A
PMV_00075	CATCACCAAGCTCGCCAACAGTT	SARS-Cov Replicase 1A

id	sequence(5'-3')	region
PMV_00076	AGGTTGCCATCATTTTGGCATCTT	SARS-Cov Replicase 1A
PMV_00077	CTTTGCGCCAGCGATAGTGACTT	SARS-Cov Replicase 1A
PMV_00078	ATGGCACCCGTTTCTGCAATGG	SARS-Cov Replicase 1A
PMV_00079	TCGGGCAGCTGACACGAATGTAGA	SARS-Cov Replicase 1A
PMV_00080	GAATGGCGATGTAGTGGCTATTGA	SARS-Cov Replicase 1A
PMV_00081	TAATGCCGGCATCCAAACATAAT	SARS-Cov Replicase 1A
PMV_00082	TAGCCAGCGTGGTGGTTCATACAA	SARS-Cov Replicase 1A
PMV_00083	CTCCCGGCAGAAAGCTGTAAGCT	SARS-Cov Replicase 1A
PMV_00084	TATAGAGCCCGTGCTGGTGATGC	SARS-Cov Replicase 1A
PMV_00085	ATCGCCATTCAAGTCTGGGAAGAA	SARS-Cov Replicase 1A
PMV_00086	TGGCTCAGGCCATACTGGCATTAC	SARS-Cov Replicase 1A
PMV_00087	TTTGCGCCAGCGATAGTGACTTG	SARS-Cov Replicase 1A
PMV_00088	TTCCCGTCAGGCAAAGTTGAAGG	SARS-Cov Replicase 1A
PMV_00089	GACGGCAATTCCTGTTTGAGCAGA	SARS-Cov Replicase 1A
PMV_00003	TCACTTGCTTCCGTTGAGGATGAATTACCAAGTCAATGGTTAC	SARS-Cov Replicase 1B
PMV_00004	GGTTTCGGATGTTACAGCGTATAACCAGTCGGTACAGCTAC	SARS-Cov Replicase 1B
PMV_00005	TCACTTGCTTCCGTTGAGGGAAGCTATTCGTCACGTTCG	SARS-Cov Replicase 1B
PMV_00006	GGTTTCGGATGTTACAGCGTCTGTAGAAAATCCTAGCTGGAG	SARS-Cov Replicase 1B
PMV_00007	TCACTTGCTTCCGTTGAGGCCTCTCTTGTTCTTGCTCGCA	SARS-Cov Replicase 1B
PMV_00008	GGTTTCGGATGTTACAGCGTGTGAGCCGCCACACATG	SARS-Cov Replicase 1B
PMV_00009	TCACTTGCTTCCGTTGAGGCTAACATGCTTAGGATAATGG	SARS-Cov Replicase 1B
PMV_00010	GGTTTCGGATGTTACAGCGTCAGGTAAGCGTAAAACTCATC	SARS-Cov Replicase 1B
PMV_00011	TCACTTGCTTCCGTTGAGGGCCTCTCTTGTTCTTGCTCGC	SARS-Cov Replicase 1B
PMV_00013	TCACTTGCTTCCGTTGAGGCACCGTTTCTACAGGTTAGCTAACGA	SARS-Cov Replicase 1B
PMV_00014	GGTTTCGGATGTTACAGCGTAAATGTTTACGCAGGTAAGCGTAAAA	SARS-Cov Replicase 1B
PMV_00015	TCACTTGCTTCCGTTGAGGTACACACCTCAGCGTTG	SARS-Cov Replicase 1B
PMV_00016	GGTTTCGGATGTTACAGCGTCACGAACGTGACGAAT	SARS-Cov Replicase 1B
PMV_00017	TCACTTGCTTCCGTTGAGGGCTTAGGATAATGGCCTCTC	SARS-Cov Replicase 1B
PMV_00018	GGTTTCGGATGTTACAGCGTCCACGAATTCATGATCAACATCCC	SARS-Cov Replicase 1B

id	sequence(5'-3')	region
PMV_00019	TCACTTGCTTCCGTTGAGGGCTCGCAAACATAACACTTGC	SARS-Cov Replicase 1B
PMV_00020	GGTTTCGGATGTTACAGCGTGAGACACTCATAGAGCCTGTG	SARS-Cov Replicase 1B
PMV_00055	ATGAATTACCAAGTCAATGGTTAC	SARS-Cov Replicase 1B
PMV_00056	ATAACCAGTCGGTACAGCTAC	SARS-Cov Replicase 1B
PMV_00057	GAAGCTATTCGTCACGTTCG	SARS-Cov Replicase 1B
PMV_00058	CTGTAGAAAATCCTAGCTGGAG	SARS-Cov Replicase 1B
PMV_00059	CCTCTCTTGTTCTTGCTCGCA	SARS-Cov Replicase 1B
PMV_00060	GTGAGCCGCCACACATG	SARS-Cov Replicase 1B
PMV_00061	CTAACATGCTTAGGATAATGG	SARS-Cov Replicase 1B
PMV_00062	CAGGTAAGCGTAAAACTCATC	SARS-Cov Replicase 1B
PMV_00063	GCCTCTCTTGTTCTTGCTCGC	SARS-Cov Replicase 1B
PMV_00064	CACCGTTTCTACAGGTTAGCTAACGA	SARS-Cov Replicase 1B
PMV_00065	AAATGTTTACGCAGGTAAGCGTAAAA	SARS-Cov Replicase 1B
PMV_00066	TACACACCTCAGCGTTG	SARS-Cov Replicase 1B
PMV_00067	CACGAACGTGACGAAT	SARS-Cov Replicase 1B
PMV_00068	GCTTAGGATAATGGCCTCTC	SARS-Cov Replicase 1B
PMV_00069	CCACGAATTCATGATCAACATCCC	SARS-Cov Replicase 1B
PMV_00070	GCTCGCAAACATAACACTTGC	SARS-Cov Replicase 1B
PMV_00071	GAGACACTCATAGAGCCTGTG	SARS-Cov Replicase 1B
PMSL_00003	CCAGCTCCAATAGGAATGTCGCACTC	SARS-Cov Spike glycoprotein gene
PMSL_00004	TCCGCAGATGTACATATTACAATCTACG	SARS-Cov Spike glycoprotein gene
PMSU_00005	TTAAATGCACCGGCCACGGTTTG	SARS-Cov Spike glycoprotein gene
PMV_000100	ATAGCGCCAGGACAAACTGGTGTT	SARS-Cov Spike glycoprotein gene
PMV_000101	TATATGCGCCAAGCTGGTGTGAGT	SARS-Cov Spike glycoprotein gene
PMV_000102	CGAGGCGGAGGTACAAATTGACAG	SARS-Cov Spike glycoprotein gene
PMV_000103	ATGAAGCCGAGCCAAACATACCAA	SARS-Cov Spike glycoprotein gene
PMV_00045	TCACTTGCTTCCGTTGAGGATGCACCGGCCACGGTTTGTG	SARS-Cov Spike glycoprotein gene
PMV_00046	GGTTTCGGATGTTACAGCGTATGCGCCAAGCTGGTGTGAGTTGA	SARS-Cov Spike glycoprotein gene
PMV_00047	TCACTTGCTTCCGTTGAGGTGCTGGCGCTGCTCTTCAAATACC	SARS-Cov Spike glycoprotein gene

id	sequence(5'-3')	region
PMV_00048	GGTTTCGGATGTTACAGCGTCGGGGCTGCTTGTGGGAAGG	SARS-Cov Spike glycoprotein gene
PMV_00049	TCACTTGCTTCCGTTGAGGATAGCGCCAGGACAAACTGGTGTT	SARS-Cov Spike glycoprotein gene
PMV_00050	GGTTTCGGATGTTACAGCGTTATATGCGCCAAGCTGGTGTGAGT	SARS-Cov Spike glycoprotein gene
PMV_00051	TCACTTGCTTCCGTTGAGGCGAGGCGGAGGTACAAATTGACAG	SARS-Cov Spike glycoprotein gene
PMV_00052	GGTTTCGGATGTTACAGCGTATGAAGCCGAGCCAAACATACCAA	SARS-Cov Spike glycoprotein gene
PMV_00096	ATGCACCGGCCACGGTTTGTG	SARS-Cov Spike glycoprotein gene
PMV_00097	ATGCGCCAAGCTGGTGTGAGTTGA	SARS-Cov Spike glycoprotein gene
PMV_00098	TGCTGGCGCTGCTCTTCAAATACC	SARS-Cov Spike glycoprotein gene
PMV_00099	CGGGGCTGCTTGTGGGAAGG	SARS-Cov Spike glycoprotein gene

In yet another example, the PCR is conducted using at least one of the following pairs of primers for a non-SARS-CoV infectious organism causing SARS-like symptoms set forth in Table 19.

Table 19. Exemplary primers for non-SARS-CoV infectious organism causing SARS-like symptoms

id	Sequence (5' - 3')	species
PMIA_00001	TTTGTGCGACAATGCTTCA	Influenza A virus
PMIA_00002	GACATTTGAGAAAGCTTGCC	Influenza A virus
PMIA_00003	AGGGACAACCTNGAACCTGG	Influenza A virus
PMIA_00004	AGGAGTTGAACCAAGACGCATT	Influenza A virus
PMIA_00005	ACCACATTCCCTTATACTGGAG	Influenza A virus
PMIA_00006	TTAGTCATCATCTTTCTCACAACA	Influenza A virus
PMIA_00007	ACAAATTGCTTCAAATGAGAAC	Influenza A virus
PMIA_00008	TGTCTCCGAAGAAATAAGATCC	Influenza A virus
PMIA_00009	GCGCAGAGACTTGAAGATGT	Influenza A virus
PMIA_00010	CCTTCCGTAGAAGGCCCT	Influenza A virus
PMIB_00001	CACAATGGCAGAATTTAGTGA	Influenza B virus
PMIB_00002	GTCAGTTTGATCCCGTAGTG	Influenza B virus

PMIB_00003	CAGATCCCAGAGTGGACTCA	Influenza B virus
PMIB_00004	TGTATTACCCAAGGGTTGTTAC	Influenza B virus
PMIB_00005	GATCAGCATGACAGTAACAGGA	Influenza B virus
PMIB_00006	ATGTTCGGTAAAAGTCGTTTAT	Influenza B virus
PMIB_00007	CCACAGGGGAGATTCCAAAG	Influenza B virus
PMIB_00008	GACATTCTTCCTGATTCATAATC	Influenza B virus
PMIB_00009	CAAACAACGGTAGACCAATATA	Influenza B virus
PMIB_00010	AGGTTCAGTATCTATCACAGTCTT	Influenza B virus
PMIB_00011	ATGTCCAACATGGATATTGAC	Influenza B virus
PMIB_00012	GCTCTTCCTATAAATCGAATG	Influenza B virus
PMIB_00013	TGATCAAGTGATCGGAAGTAG	Influenza B virus
PMIB_00014	GATGGTCTGCTTAATTGGAA	Influenza B virus
PMIB_00015	ACAGAAGATGGAGAAGGCAA	Influenza B virus
PMIB_00016	ATTGTTTCTTTGGCCTGGAT	Influenza B virus
PMAd1_00001	TGGCGGTATAGGGGTAACTG	Human adenovirus
PMAd1_00002	ATTGCGGTGATGGTTAAAGG	Human adenovirus
PMAd1_00003	TTTTGCCGATCCCACTTATC	Human adenovirus
PMAd1_00004	GCAAGTCTACCACGGCATTT	Human adenovirus
PMAd2_00001	CTCCGTTATCGCTCCATGTT	Human adenovirus
PMAd2_00002	AAGGACTGGTCGTTGGTGTC	Human adenovirus
PMAd2_00003	AAATGCCGTGGTAGATTTGC	Human adenovirus
PMAd2_00004	GTTGAAGGGGTTGACGTTGT	Human adenovirus
PMAd3_00001	TCCTCTGGATGGCATAGGAC	Human adenovirus
PMAd3_00002	TGTTGGTGTTAGTGGGCAAA	Human adenovirus
PMAd3_00003	ACATGGTCCTGCAAAGTTCC	Human adenovirus
PMAd3_00004	GCATTGTGCCACGTTGTATC	Human adenovirus
PMAd4_00001	CGCTTCGGAGTACCTCAGTC	Human adenovirus
PMAd4_00002	CTGCATCATTGGTGTCAACC	Human adenovirus
PMAd4_00003	GGCACCTTTTACCTCAACCA	Human adenovirus
PMAd4_00004	TCTGGACCAAGAACCAGTCC	Human adenovirus

PMAd5_00001	GGCCTACCCTGCTAACTTCC	Human adenovirus
PMAd5_00002	ATAAAGAAGGGTGGGCTCGT	Human adenovirus
PMAd5_00003	ATCGCAGTTGAATGCTGTTG	Human adenovirus
PMAd5_00004	GTTGAAGGGGTTGACGTTGT	Human adenovirus
PMAd7_00001	ACATGGTCCTGCAAAGTTCC	Human adenovirus
PMAd7_00002	GATCGAACCCTGATCCAAGA	Human adenovirus
PMAd7_00003	AACACCAACCGAAGGAGATG	Human adenovirus
PMAd7_00004	CCTATGCCATCCAGAGGAAA	Human adenovirus
PMAd11_00001	CAGATGCTCGCCAACTACAA	Human adenovirus
PMAd11_00002	AGCCATGTAACCCACAAAGC	Human adenovirus
PMAd11_00003	ACGGACGTTATGTGCCTTTC	Human adenovirus
PMAd11_00004	GGGAATATTGGTTGCATTGG	Human adenovirus
PMAd21_00001	ACTGGTTCCTGGTCCAGATG	Human adenovirus
PMAd21_00002	AGCCATGTAACCCACAAAGC	Human adenovirus
PMAd21_00003	CTGGATATGGCCAGCACTTT	Human adenovirus
PMAd21_00004	CACCTGAGGTTCTGGTTGGT	Human adenovirus
PMAd23_00001	TAATGAAAAGGGCGGACAAG	Human adenovirus
PMAd23_00002	GCCAATGTAGTTTGGCCTGT	Human adenovirus
PMAd23_00003	AACTCCGCGGTAGACAGCTA	Human adenovirus
PMAd23_00004	CGTAGGTGTTGGTGTTGGTG	Human adenovirus
PMV_a0061	TCACTTGCTTCCGTTGAGGTTGGGGTGA TGGGTTTCAGATTAA	HCoV-OC43
PMV_a0062	GGTTTCGGATGTTACAGCGTCTCGGGAA GATCGCCTTCTTCTA	HCoV-OC43
PMV_b0061	TTGGGGTGATGGGTTTCAGATTAA	HCoV-OC43
PMV_b0062	CTCGGGAAGATCGCCTTCTTCTA	HCoV-OC43
PMV_a0053	TCACTTGCTTCCGTTGAGGTTGGGCTGG CGGTTTAGAGTTGA	HCoV-229E
PMV_a0054	GGTTTCGGATGTTACAGCGTGTGCGACC GCCCTTGTTTATGG	HCoV-229E

PMV_a0055	TCACTTGCTTCCGTTGAGGGCGTTGTTG GCCTTTTTCTTGTCT	HCoV-229E
PMV_a0056	GGTTTCGGATGTTACAGCGTGCCCGGC ATTATTTCATTGTTCTG	HCoV-229E
PMV_a0057	TCACTTGCTTCCGTTGAGGACAAAAGCC GCTGGTGGTAAAG	HCoV-229E
PMV_a0058	GGTTTCGGATGTTACAGCGTCAGAAATC ATAACGGGCAAACTCA	HCoV-229E
PMV_a0059	TCACTTGCTTCCGTTGAGGAAGAGTTAT TGCTGGCGTTGTTGG	HCoV-229E
PMV_a0060	GGTTTCGGATGTTACAGCGTGCCCGGC ATTATTTCATTGTTCTG	HCoV-229E
PMV_b0053	TTGGGCTGGCGGTTTAGAGTTGA	HCoV-229E
PMV_b0054	GTGCGACCGCCCTTGTTTATGG	HCoV-229E
PMV_b0055	GCGTTGTTGGCCTTTTTCTTGTCT	HCoV-229E
PMV_b0056	GCCCGGCATTATTTCATTGTTCTG	HCoV-229E
PMV_b0057	ACAAAAGCCGCTGGTGGTAAAG	HCoV-229E
PMV_b0058	CAGAAATCATAACGGGCAAACTCA	HCoV-229E
PMV_b0059	AAGAGTTATTGCTGGCGTTGTTGG	HCoV-229E
PMV_b0060	GCCCGGCATTATTTCATTGTTCTG	HCoV-229E
PMHE_00001	GGTGGTAACCCCTCGCAGGA	Human enteric coronaviruse
PMHE_00002	TGGCTCTTCCCTTTGGGCACT	Human enteric coronaviruse
PMHE_00003	GAGAATGAACCTTATGTCGGCACCTG	Human enteric coronaviruse
PMHE_00004	TTCCGCAAGTCTTTCACTTTCTCCAA	Human enteric coronaviruse
PMHE_00005	CAGCTTTCAGCCAGGGACGTGT	Human enteric coronaviruse
PMHE_00006	TTTCCAGCTTTTGCGCAGTGGT	Human enteric coronaviruse
PMHE_00007	TCTGTTTTGGTGCAGGTCAATTTGTG	Human enteric coronaviruse
PMHE_00008	ATGAACCAGGTCGTAAGCATCCTCAA	Human enteric coronaviruse
PMHE_00009	GTTGCTTGTCAACCCCCGTACTGTTA	Human enteric coronaviruse
PMHE_00010	AGGACACCTGCCATAGGGGTAGAGAG	Human enteric coronaviruse

PMHE_00011	GGTTGTTGACTCGCGGTGGA	Human enteric coronaviruse
PMHE_00012	GGGGTAGAGAGGCCAAACACTGC	Human enteric coronaviruse
PMRh_00001	ACATGGTCCCATTGGATTGT	Human rhinovirus
PMRh_00002	TGAGGAAATCTTTCGCCACT	Human rhinovirus
PMRh_00003	ATGTTGCCCCCTAGTCTGTG	Human rhinovirus
PMRh_00004	TTCTGAAGGTGGTGTGTTGC	Human rhinovirus
PMRh_00005	TGGTATTCATGTTGGCGGTA	Human rhinovirus
PMRh_00006	ACAGCAGGTTCCTTGTCACC	Human rhinovirus
PMRh_00007	TCTTGCCTCCAATGGCTAGT	Human rhinovirus
PMRh_00008	TGACATGCCTGCATTGAGTT	Human rhinovirus
PMRh_00009	TCCCAATATGCCCTCTTCAG	Human rhinovirus
PMRh_00010	CGCTGATGGGGATTGAGTAT	Human rhinovirus
PMRh_00011	TGTGCTCAGTGTGCTTCCTC	Human rhinovirus
PMRh_00012	TGCACCCATGATGACAATCT	Human rhinovirus
PMRh_00013	GCAGTTCTTGCCAAAGAAGG	Human rhinovirus
PMRh_00014	TGAAGGGTTTTTGGTCCATC	Human rhinovirus
PMRh_00015	TGCCTGATGCCCTTAAAAAC	Human rhinovirus
PMRh_00016	GGGTGTGATTGTACCCGACT	Human rhinovirus
PMMP_00001	CTTAACAGTTGTATGCATTGGAAACT	Mycoplasma pneumoniae
PMMP_00002	GTTTACGGTGTGGACTACTAGGGTAT	Mycoplasma pneumoniae
PMMP_00003	CTATGCTGAGAAGTAGAATAGCCACA	Mycoplasma pneumoniae
PMMP_00004	TGGTACAGTCAAACTCTAGCCATTAC	Mycoplasma pneumoniae
PMMP_00005	ATACCCTAGTAGTCCACACCGTAAAC	Mycoplasma pneumoniae
PMMP_00006	ATGTCAAGTCTAGGTAAGGTTTTTCG	Mycoplasma pneumoniae
PMMP_00007	AGGCGAAAACTTAGGCCATT	Mycoplasma pneumoniae
PMMP_00008	CCGTCAATTCCGTTTGAGTT	Mycoplasma pneumoniae
PMMP_00009	CGACGGTACACGAAAAACCT	Mycoplasma pneumoniae
PMMP_00010	TCCCTTCCTTCCTCCAATTT	Mycoplasma pneumoniae
PMR_00001	ATTCCCATGGAGAAACTCCTAGAT	Rubella virus
PMR_00002	GTGATCACTGACCTGCATCTG	Rubella virus

PMR_00003	GTAAGAGACCACGTCCGATCAAT	Rubella virus
PMR_00004	GAGGACGTGTAGGGCTTCTTTAG	Rubella virus
PMR_00005	ATCGGACCTCGCTTAGGACT	Rubella virus
PMR_00006	CTGGGTATCACGGCTACGAT	Rubella virus
PMR_00007	AGAGACCACGTCCGATCAAT	Rubella virus
PMR_00008	TGAGGACGTGTAGGGCTTCT	Rubella virus
PMR_00009	GTCAACGCCTACTCCTCTGG	Rubella virus
PMR_00010	GTCTTGTGAGGGTGCTGGAC	Rubella virus
PMM_00001	CACATTGGCATCTGAACTCG	Measles virus
PMM_00002	TCTGTTTGACCCTCCTGTCC	Measles virus
PMM_00003	AGATTGCAATGCATACTACTGAGGAC	Measles virus
PMM_00004	ATGCAGTGTCAATGTCTAGAGGTGT	Measles virus
PMM_00005	CAATGCATACTACTGAGGACAGGA	Measles virus
PMM_00006	ATGCAGTGTCAATGTCTAGAGGTG	Measles virus
PMM_00007	TACCATCAGAGGTCAATTCTCAAA	Measles virus
PMM_00008	CTACTTCAAACACTCGGTACATGC	Measles virus
PMM_00009	CATGTCGCTGTCTCTGTTAGACTT	Measles virus
PMM_00010	CAAGCCTGGATTTCTTATAACACC	Measles virus
PMRSV_00001	AAACCAAAGAAGAAACCAACCAT	Human respiratory syncytial virus
PMRSV_00002	TGTTCTAATGTGGTTGTGTCGAG	Human respiratory syncytial virus
PMRSV_00003	TGCTAAAAGAGATGGGAGAAGTG	Human respiratory syncytial virus
PMRSV_00004	ATCCTTTGGTATGAGACCCTTGT	Human respiratory syncytial virus
PMRSV_00005	ACAAGGGTCTCATACCAAAGGAT	Human respiratory syncytial virus
PMRSV_00006	GCTAAAACTCCCCATCTTAGCAT	Human respiratory syncytial virus

PMRSV_00007	TTTATGATGCAGCCAAAGCA	Human respiratory syncytial virus
PMRSV_00008	TCCATGAAATTCAGGTGCAA	Human respiratory syncytial virus
PMRSV_00009	AAAAACACCAGCCAAAACGA	Human respiratory syncytial virus
PMRSV_00010	CTGTGGGTGTTTGTGTGGAG	Human respiratory syncytial virus
PMRSV_00011	CCAAAGCATATGCAGAGCAA	Human respiratory syncytial virus
PMRSV_00012	TCCATGAAATTCAGGTGCAA	Human respiratory syncytial virus
PMPI_00001	GCATGGAAACTAGCAGCACA	Parainfluenza
PMPI_00002	GGTGTTGTGGTCTTCGAGGT	Parainfluenza
PMPI_00003	GGCTCCATAGTATCATCGACAAC	Parainfluenza
PMPI_00004	CCTAGAGGCCCTGTGTATACCTT	Parainfluenza
PMPI_00005	ACACAACAAACAATGCAAACAAC	Parainfluenza
PMPI_00006	TTAACATGCGCTTAGCAAATACA	Parainfluenza
PMPI_00007	TTAGCTCACTCATTGGACACAGA	Parainfluenza
PMPI_00008	GTCTCTCGTTTTGACAATGAACC	Parainfluenza
PMPI_00009	TCTCACTACAAACGGTGTCAATG	Parainfluenza
PMPI_00010	TCTAGATCCGCATTCTCTCTTTG	Parainfluenza
PMPI_00011	ACAGATGGGTTCATTGTCAAAAC	Parainfluenza
PMPI_00012	GCTTTGACCAACACTATCCAAAC	Parainfluenza
PMPI_00013	GCTGAACACCCAGATTTACAAAG	Parainfluenza
PMPI_00014	ACAGCTCTCCATTTCATGGTTTA	Parainfluenza
PMPI_00015	ATATGCATTTGTCAATGGAGGAG	Parainfluenza
PMPI_00016	CATTTGGTGTGTAAAATGCAAGA	Parainfluenza
PMPI_00017	CACAGAACACCAGAACAACAAGA	Parainfluenza
PMPI_00018	TTGGGACTGTTAACCAATACACC	Parainfluenza

PMME_00001	CATCCCAAAAATTGCCAGAT	Human metapneumovirus
PMME_00002	TTTGGGCTTTGCCTTAAATG	Human metapneumovirus
PMME_00003	ACACCCTCATCATTGCAACA	Human metapneumovirus
PMME_00004	GCCCTTCTGACTGTGGTCTC	Human metapneumovirus
PMME_00005	CGACACAGCAGCAGGAATTA	Human metapneumovirus
PMME_00006	TCAAAGCTGCTTGACACTGG	Human metapneumovirus

In yet another example, the PCR is conducted using at least one of the following pairs of primers for a non-SARS-CoV infectious organism damaging the subject's immune system set forth in Table 20.

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Table 20. Exemplary primers for non-SARS-CoV infectious organism damaging the subject's immune system

id	sequence(5' - 3')	species
PMTTV_00001	TGGGGCCAGACTTCGCCATA	TTV
PMTTV_00002	AGCTTCCGCCGAGGATGACC	TTV
PMTTV_00003	CTTGGGGGCTCAACGCCTTC	TTV
PMTTV_00004	GCGAAGTCTGGCCCCACTCA	TTV
PMTTV_00005	CCACAGGCCAACCGAATGCT	TTV
PMTTV_00006	AGCCCGAATTGCCCCTTGAC	TTV
PMTTV_00007	AGCGAATCCTGGGAGTCAAACTCAG	TTV
PMTTV_00008	GGCCTCGTACTCCTCTTTCCAGTCA	TTV
PMTTV_00009	GCCCCTTTGCATACCACTCAGACAT	TTV
PMTTV_00010	TGGAATGTGAGTTCCGGTGAGTTGT	TTV
PMTTV_00011	TGTCAGTAACAGGGGTCGCCATAGA	TTV
PMTTV_00012	TGTGACGTATGGACGACCTTTGACC	TTV
PMV_11047	CACAGACAGAGGAGAAGGCAAC	TTV
PMV_11048	AATAGGCACATTACTACTACCTCCTG	TTV
PMTP_00001	GCGGTCGGTAGGAGGATAAAGGAAA	TP
PMTP_00002	CCGGGGATTTGTCTACAGGGTTTCT	TP

PMTP_00003	CAGACGCTCATCCAACTCCTGAGAA	TP
PMTP_00004	CCGTTGTACCGTCTTTTTGGACGTT	TP
PMTP_00005	CACGCTCTACCTCATTCGAGAGCAA	TP
PMTP_00006	GTTGTGTTGCAACGAACACGCTACA	TP
PMTP_00007	AGCGGTCGGTAGGAGGATAAAGGAA	TP
PMTP_00008	ACCGGGGATTTGTCTACAGGGTTTC	TP
PMV_11025	AACACGATCCGCTACGACTACTAC	TP
PMV_11026	CCCTATACCCGTTCGCAATCAAAG	TP
PMHIV1_00001	ATGGGCGCAGCCTCAATGAC	HIV1
PMHIV1_00002	CCCCAAATCCCCAGGAGCTG	HIV1
PMHIV1_00003	GGGACAGCTACAACCATCCCTTCAG	HIV1
PMHIV1_00004	GACCTGATTGCTGTGTCCTGTGTCA	HIV1
PMHIV1_00005	GGGATGGAAAGGATCACCAGCAATA	HIV1
PMHIV1_00006	GTCTGGTGTGGTAAGTCCCCACCTC	HIV1
PMHIV1_00007	AAGGATCAACAGCTCCTGGGGATTT	HIV1
PMHIV1_00008	TTCTTGCTGGTTTTGCGATTCTTCA	HIV1
PMV_11055	TAATCCACCTATCCCAGTAGGAGAAAT	HIV1
PMV_11056	GGTCCTTGTCTTATGTCCAGAATGC	HIV1
PMV_11057	TGGGAAGTTCAATTAGGAATACCAC	HIV1
PMV_11058	TCCTACATACAAATCATCCATGTATTG	HIV1
PMHGV_00001	GCCGGCGATGACTGCTTGAT	HGV
PMHGV_00002	TCCGGAAGTCCGTGGTCAGG	HGV
PMHGV_00003	ACGGTGGGAGTCGCGTTGAC	HGV
PMHGV_00004	GGCCACGCAAACCAACAAGG	HGV
PMHGV_00005	CGGCCAAAAGGTGGTGGATG	HGV
PMHGV_00006	CGGGCTCGGTTTAACGACGA	HGV
PMHGV_00007	GCCACGGGCAAAATCAGTGG	HGV
PMHGV_00008	TGTCGCGATCCGATGATCCA	HGV
PMHGV_00009	CGCGTGTGAGCTAAAGTGGGAAAGT	HGV
PMHGV_00010	ATCGTCACCAACAGGAAGACCATGA	HGV

PMHGV_00011	TCGCTCTCGGGTTGGTTTTGTATTC	HGV
PMHGV_00012	CATCCACCTTAGGCTCCCTGTTGAC	HGV
PMV_11045	GGGTTGGTAGGTCGTAAATCCC	HGV
PMV_11046	GTACGTGGGCGTCGTTTGC	HGV
PMV_11001	CCTTTCCACCATCCAGCAGT	HEV
PMV_11002	CGAGCTTTACCCACCTTCAGC	HEV
PMHEV_00001	CTGGCGGTGGGCTCTGTCAT	HEV
PMHEV_00002	ACCGAGGCGGGAGCAAGTCT	HEV
PMHEV_00003	ACGGGCGGATCGATTGTGAG	HEV
PMHEV_00004	GGCAGCGACATAGCGCACCT	HEV
PMHEV_00005	AGCTCACCACCACGGCTGCT	HEV
PMHEV_00006	CTGAGACGACGGGGCGAGAG	HEV
PMHEV_00007	ATCGCGCCCCTTTTCTGTCC	HEV
PMHEV_00008	GGGGGCGACCATCAAGTGTG	HEV
PMHDV_00001	GACGGGCCGGCTGTTCTTCT	HDV
PMHDV_00002	GACTCCGGGCCTGGGAAGAG	HDV
PMHDV_00003	ACTCCGGCCGAAAGGTCGAG	HDV
PMHDV_00004	GGCGGAACACCCACCGACTA	HDV
PMHDV_00005	CCATGACTCTGGAGACATCCTGGAA	HDV
PMHDV_00006	CGTCAGAGCTCTCTGTTCGCTGAAG	HDV
PMHDV_00007	CCTTCTCTCGTCTTCCTCGGTCAAC	HDV
PMHDV_00008	CCGAACGGACCAGATGGAGATAGAC	HDV
PMHDV_00009	GCTCCCGAGAGGGATAAAACGGTAA	HDV
PMHDV_00010	GAGTGCTCTCCAAACTTGGCAGTTG	HDV
PMHDV_00011	TCTCGTCTTCCTCGGTCAACCTCTT	HDV
PMHDV_00012	CCGAACGGACCAGATGGAGATAGAC	HDV
PMV_11041	AACATTCCGAAGGGGACCGT	HDV
PMV_11042	GGCATCCGAAGGAGGACG	HDV
PMHCV_00001	GGCGCTGGAAAGAGGGTCTACTACC	HCV
PMHCV_00002	TGTTCAAGCTGATCCCTGGCTATGA	HCV

PMHCV_00003	ACATCTGGGACTGGATATGCGAGGT	HCV
PMHCV_00004	ATCCTCATCGTCCCGTTTTTGACAT	HCV
PMHCV_00005	TGTGCCAGGACCATCTTGAATTTTG	HCV
PMHCV_00006	AGGCGGATCAAACACTTCCACATCT	HCV
PMHCV_00007	GGGGTGCAAATGATACGGATGTCTT	HCV
PMHCV_00008	AGAGTATGTGGCTTCCGGATGCTTG	HCV
PMHCV_00009	ACACGCCGTGGGCCTATTCA	HCV
PMHCV_00010	GCCGGGACCTTGGTGCTCTT	HCV
PMHCV_00011	CACGCCGTGGGCCTATTCAG	HCV
PMHCV_00012	GCCGGGACCTTGGTGCTCTT	HCV
PMV_11039	CTCGCAAGCACCCTATCAGGCAGT	HCV
PMV_11040	GCAGAAAGCGTCTAGCCATGGCGT	HCV
PMHCMV_00001	GCGCCTGCTGCTCGAAATGT	HCMV
PMHCMV_00002	GTCGCGGCTGTTGCGGTAGT	HCMV
PMHCMV_00003	CCCCACGTCCATCTGCGTCT	HCMV
PMHCMV_00004	GCCCCCAGCAGTCTCACCAG	HCMV
PMHCMV_00005	GCTCACGCACCCTGGAGGAC	HCMV
PMHCMV_00006	AGTTCCAGCCCACGCACCAG	HCMV
PMHCMV_00007	GTGCAGTTTAGGTGGCAGTTCATGC	HCMV
PMHCMV_00008	GGAAAGGGGAGGGTAGAAACGTGAG	HCMV
PMHCMV_00009	TGTGATTGCGTGTGCAGTTTAGGTG	HCMV
PMHCMV_00010	GGGGAGGGTAGAAACGTGAGTCTCC	HCMV
PMV_11051	ATTCCAAGCGGCCTCTGATAA	HCMV
PMV_11052	TCTTCCTCTGGGGCAACTTCC	HCMV
PMHBV_00001	TCGCAGTCCCCAACCTCCAA	HBV
PMHBV_00002	CAGGGTCCCGTGCTGGTTGT	HBV
PMHBV_00003	GCAGCCGGTCTGGAGCAAAA	HBV
PMHBV_00004	GCAGACGGAGAAGGGGACGA	HBV
PMHBV_00005	CGCCTCATTTTGCGGGTCAC	HBV
PMHBV_00006	TGGTTGGCTTGTGGCCAGTG	HBV

PMHBV_00007	ATCAAGGTATGTTGCCCGTTTGTCC	HBV
PMHBV_00008	AGGCCCACTCCCATAGGTATTTTGC	HBV
PMHBV_00009	CCTAGGACCCCTGCTCGTGTTACAG	HBV
PMHBV_00010	GCGATAACCAGGACAAATTGGAGGA	HBV
PMHBV_00011	CTGCGCACCATTATCATGCAACTTT	HBV
PMHBV_00012	AGTAGATCCCGGACGGAAGGAAAGA	HBV
PMV_11037	GTTCAAGCCTCCAAGCTGTG	HBV
PMV_11038	TCAGAAGGCAAAAAAGAGAGTAACT	HBV
PMHAV_00001	GATGTTTGGGACGTCACCTT	HAV
PMHAV_00002	CTGGATGAGAGCCAGTCCTC	HAV
PMHAV_00003	ATTGCATTGGCAACCAAAAT	HAV
PMHAV_00004	ATCTCATTGGGCATCCTGAC	HAV
PMHAV_00005	GACTGGAGGTTGGGAAACAA	HAV
PMHAV_00006	AGCAGCCAGAGAGAATCCAA	HAV
PMHAV_00007	TAAGCATTTTTCCCGCAAAG	HAV
PMHAV_00008	AGGCATTCATGACCCATCTC	HAV
PMHAV_00009	CCAACCAAATATCATTCAGGTAGAC	HAV
PMHAV_00010	GACTTCGTGTACCTATTCACTCGAT	HAV
PMHAV_00011	GGGTTTCCTTATGTTCAAGAAAAAT	HAV
PMHAV_00012	CCAAAACTTTCTCTAATGGTCTCAA	HAV
PMV_11035	TTTTGCTCCTCTTTACCATGCTATG	HAV
PMV_11036	GGAAATGTCTCAGGTACTTTCTTTG	HAV
PMEBV_00001	AACCCAATAGCATGACAGCCAATCC	EBV
PMEBV_00002	TCAGCCCCAGAGACACGGTATATGA	EBV
PMEBV_00003	TGAACCTGGGACCTATTGATGCAGA	EBV
PMEBV_00004	CAGGGGAATCTCTGCCAACTTTGAG	EBV
PMEBV_00005	TGCACAGTGACAGTGGGAGAAACAC	EBV
PMEBV_00006	AAGAATGGAAAGGGTTGGCAGTGTG	EBV
PMEBV_00007	GTGCACAGTGACAGTGGGAGAAACA	EBV
PMEBV_00008	AAGAATGGAAAGGGTTGGCAGTGTG	EBV

PMV_11053	CCCACGCGCGCATAATG	EBV
PMV_11054	TTCACTTCGGTCTCCCCTAG	EBV
PMB19_00001	TGGGCCGCCAAGTACAGGAA	B19
PMB19_00002	GGGTTGCCCGCCTAAAATGG	B19
PMB19_00003	CCCTATTAGTGGGGCAGCATGTGTT	B19
PMB19_00004	CCACCAAGCTTTTCCCTGCTACATC	B19
PMB19_00005	CAGTGTCACAGCCATACCACCACTG	B19
PMB19_00006	TGCTGGGTTCCTTTATTGGGGAAAT	B19
PMB19_00007	CCCATTGCATTAATGTAGGGGCTTG	B19
PMB19_00008	ATCACTTTCCCACCATTTGCCACTT	B19
PMV_11049	CCTTTCCACCATCCAGCAGT	B19
PMV_11050	CGAGCTTTACCCACCTTCAGC	B19

In yet another example, the PCR is conducted using at least one of the following pairs of primers for a non-SARS-CoV coronaviridae virus set forth in Table 21.

Table 21. Exemplary primers for non-SARS-CoV coronaviridae virus

seqid	sequence(5'-3')
PMIBV_00001	GGAACAGGACCTGCCGCTGA
PMIBV_00002	ATCAGGTCCGCCATCCGAGA
PMIBV_00003	AAAGGTGGAAGAAAACCAGTCCCAGA
PMIBV_00004	GCCATCCGAGAATCGTAGTGGGTATT
PMMHV_00001	CAGCGCCAGCCTGCCTCTAC
PMMHV_00002	TGCTGCACTGGGCACTGCTT
PMMHV_00003	GGAAATTACCGACTGCCCTCAAACA
PMMHV_00004	TGATTATTTGGTCCACGCTCGGTTT
PMEQ_00001	TCCCGCGCATCCAGTAGAGC
PMEQ_00002	CTGCGGCTTTGTGGCATCCT
PMEQ_00003	TTTGCTGAAGGACAAGGTGTGCCTA

PMEQ_00004	CCAGAAGACTCCGTCAATGTTGGTG
PMCA_00001	AAAAACGTGGTCGTTCCAATTCTCG
PMCA_00002	CCATGCGATAGCGGCTTTGTCTATT
PMCA_00003	TGGGAACGGTGCCAAGCATT
PMCA_00004	GCCACCTCTGATGGACGAGCA
PMFE_00001	CGCGTCAACTGGGGAGATGAA
PMFE_00002	GCGCGCCTGTCTGTTCCAAT
PMFE_00003	GAGTCTTCTGGGTTGCAAAGGATGG
PMFE_00004	CCCCTGGATTGAGACCTGTTTCTTG
PMPEDV_00001	GCAGCATTGCTCTTTGGTGGTAATG
PMPEDV_00002	TGCTGAATGGTTTCACGCTTGTTCT
PMPEDV_00003	CCGCAAACGGGTGCCATTAT
PMPEDV_00004	TCGCCGTGAGGTCCTGTTCC
PMPTGV_00001	TCGCTCCAATTCCCGTGGTC
PMPTGV_00002	ACGTTGGCCCTTCACCATGC
PMPTGV_00003	CAAGCATTACCCACAATTGGCTGAA
PMPTGV_00004	TTCTTTTGCCACTTCTGATGGACGA
PMBOV_00001	TTCCTTTAAAACAGCCGATGGCAAC
PMBOV_00002	TCGGAATAGCCTCATCGCTACTTGG
PMBOV_00003	TTCCGCCTGGCACGGTACTC
PMBOV_00004	TGGCTTAGCGGCATCCTTGC
PMFIPV_00001	CACCATGGCCTCAGCCTTGA
PMFIPV_00002	GTGCCGCCAACCTGCCAGTA
PMFIPV_00003	GGTCTTGGCACTGTGGATGATGATT
PMFIPV_00004	GAAAAAGGGACAGCTACAGCGGATG
PMR_00001	CCCAATCAGAATTTTGGAGGCTCTG
PMR_00002	AGCGAATTGCACCTGAATACTGCAA
PMR_00003	TGACCAAACCGAGCGTGCAG
PMR_00004	CAGTGGCGGGGATTCCATTG
PMPHEV_00001	AGCGTCAACTGCTGCCACGA

PMPHEV_00002	AGTACCGTGCCAGGCGGAAA
PMPHEV_00003	AAGGTGTGCCTATTGCACCAGGAGT
PMPHEV_00004	ACTAGCGACCCAGAAGACTCCGTCA
PMPV_00001	AGAAGACCACTTGGGCTGACCAAAC
PMPV_00002	TTGGCAATAGGCACTCCTTGTCCTT
PMPV_00003	GCGCCAGCCTGCCTCTATTG
PMPV_00004	TGGGGCCCCTCTTTCCAAAA
PMTK_00001	ATGGCTCACCGCCGGTATTG
PMTK_00002	TGGGCGTCACTCTGCTTCCA
PMTK_00003	GCTAAGGCTGATGAAATGGCTCACC
PMTK_00004	TCCAAAAAGACAAGCATGGCTGCTA
PMSDAV_00001	TCTATGTTGAAGGCTCGGGAAGGTC
PMSDAV_00002	TACTTGCTTAGGCTGTCCGGCATCT
PMSDAV_00003	AGCAGTGCCCAGTGCAGCAG
PMSDAV_00004	TGGGTTCATCAACGCCACCA

D. SARS-CoV and non-SARS-CoV infectious organism primers, probes, kit and uses thereof

In still another aspect, the present invention is directed to an oligonucleotide primer for amplifying a SARS-CoV and/or a non-SARS-CoV infectious organism nucleotide sequence, which oligonucleotide primer comprises a nucleotide sequence that: a) hybridizes, under high stringency, with a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence, or a complementary strand thereof, that is set forth in Table 18 or Tables 19-21; or b) has at least 90% identity to a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 18 or Tables 19-21.

The present primers can comprise any suitable types of nucleic acids, e.g., DNA, RNA, PNA or a derivative thereof. Preferably, the primers comprise a nucleotide

sequence, or a complementary strand thereof, that is set forth in Table 18 or Tables 19-21.

In a specific embodiment, the present invention is directed to a kit for amplifying a SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence, which kit comprises: a) an above-described primer; and b) a nucleic acid polymerase that can amplify a SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence using the probe. Preferably, the nucleic acid polymerase is a reverse transcriptase.

In yet another aspect, the present invention is directed to an oligonucleotide probe for hybridizing to a SARS-CoV or a non-SARS-CoV infectious organismnucleotide sequence, which oligonucleotide probe comprises a nucleotide sequence that: a) hybridizes, under high stringency, with a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17; or b) has at least 90% identity to a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence comprising a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17.

The present probes can comprise any suitable types of nucleic acids, e.g., DNA, RNA, PNA or a derivative thereof. Preferably, the probes comprise a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17. Also preferably, the probes are labeled, e.g., a chemical, an enzymatic, an immunogenic, a radioactive, a fluorescent, a luminescent and a FRET label.

In a specific embodiment, the present invention is directed to a kit for hybridization analysis of a SARS-CoV and/or a non-SARS-CoV infectious organism nucleotide sequence, which kit comprises: a) an above-described probe; and b) a means for assessing a hybrid formed between a SARS-CoV and/or a non-SARS-CoV infectious organism nucleotide sequence and said probe.

The oligonucleotide primers and probes can be produced by any suitable method. For example, the probes can be chemically synthesized (See generally, Ausubel (Ed.) Current Protocols in Molecular Biology, 2.11. Synthesis and purification of oligonucleotides, John Wiley & Sons, Inc. (2000)), isolated from a natural source,

produced by recombinant methods or a combination thereof. Synthetic oligonucleotides can also be prepared by using the triester method of Matteucci et al., J. Am. Chem. Soc., 3:3185-3191 (1981). Alternatively, automated synthesis may be preferred, for example, on a Applied Biosynthesis DNA synthesizer using cyanoethyl phosphoramidite chemistry. Preferably, the probes and the primers are chemically synthesized.

Suitable bases for preparing the oligonucleotide probes and primers of the present invention may be selected from naturally occurring nucleotide bases such as adenine, cytosine, guanine, uracil, and thymine. It may also be selected from nonnaturally occurring or "synthetic" nucleotide bases such as 8-oxo-guanine, 6-mercaptoguanine, 4-acetylcytidine, 5-(carboxyhydroxyethyl) uridine, 2'-O-methylcytidine, 5-carboxymethylamino-methyl-2-thioridine, 5-carboxymethylaminomethyl uridine, dihydrouridine, 2'-O-methylpseudouridine, beta-D-galactosylqueosine, 2'-Omethylguanosine, inosine, N6 -isopentenyladenosine, 1-methyladenosine, 1-methylpseudouridine, 1-methylguanosine, 1-methylinosine, 2,2-dimethylguanosine, 2-methyladenosine, 2-methylguanosine, 3-methylcytidine, 5-methylcytidine, N6 -methyladenosine, 7-methylguanosine, 5-methylaminomethyluridine, 5-methoxyaminomethyl-2-thiouridine, beta-D-mannosylqueosine, 5-methoxycarbonylmethyluridine, 5-methoxyuridine, 2-methylthio-N6 -isopentenyladenosine, N-((9-.beta.-D-ribofuranosyl-2-methylthiopurine-6-yl)carbamoyl)threonine, N-((9-beta-D-ribofuranosylpurine-6-yl) N-methylcarbamoyl) threonine, uridine-5-oxyacetic acid methylester, uridine-5-oxyacetic acid, wybutoxosine, pseudouridine, queosine, 2-thiocytidine, 5-methyl-2-thiouridine, 2-thiouridine, 2-thiouridine, 5-methyluridine, N-((9-beta-D-ribofuranosylpurine-6-yl) carbamoyl) threonine, 2'-O-methyl-5-methyluridine, 2'-O-methyluridine, wybutosine, and 3-(3-amino-3-carboxypropyl) uridine.

Likewise, chemical analogs of oligonucleotides (e.g., oligonucleotides in which the phosphodiester bonds have been modified, e.g., to the methylphosphonate, the phosphotriester, the phosphorothioate, the phosphorodithioate, or the phosphoramidate) may also be employed. Protection from degradation can be achieved by use of a "3'-end

cap" strategy by which nuclease-resistant linkages are substituted for phosphodiester linkages at the 3' end of the oligonucleotide (Shaw et al., Nucleic Acids Res., 19:747 (1991)). Phosphoramidates, phosphorothioates, and methylphosphonate linkages all function adequately in this manner. More extensive modification of the phosphodiester backbone has been shown to impart stability and may allow for enhanced affinity and increased cellular permeation of oligonucleotides (Milligan et al., J. Med. Chem., 36:1923 (1993)). Many different chemical strategies have been employed to replace the entire phosphodiester backbone with novel linkages. Backbone analogues include phosphorothioate, phosphorodithioate, methylphosphonate, phosphoramidate, boranophosphate, phosphotriester, formacetal, 3 '-thioformacetal, 5'-thioformacetal, 5'-thioether, carbonate, 5'-N-carbamate, sulfate, sulfonate, sulfamate, sulfonamide, sulfone, sulfite, sulfoxide, sulfide, hydroxylamine, methylene (methylimino) (MMI) or methyleneoxy (methylimino) (MOMI) linkages. Phosphorothioate and methylphosphonate-modified oligonucleotides are particularly preferred due to their availability through automated oligonucleotide synthesis. The oligonucleotide may be a "peptide nucleic acid" such as described by (Milligan et al., J. Med. Chem., 36:1923 (1993)). The only requirement is that the oligonucleotide probe should possess a sequence at least a portion of which is capable of binding to a portion of the sequence of a target SARS-CoV sequence.

Hybridization probes or amplification primers can be of any suitable length. There is no lower or upper limits to the length of the probe or primer, as long as the probe hybridizes to the SARS-CoV or the non-SARS-CoV infectious organism target nucleic acids and functions effectively as a probe or primer (e.g., facilitates detection or amplification). The probes and primers of the present invention can be as short as 50, 40, 30, 20, 15, or 10 nucleotides, or shorter. Likewise, the probes or primers can be as long as 20, 40, 50, 60, 75, 100 or 200 nucleotides, or longer, e.g., to the full length of the SARS-CoV or the non-SARS-CoV infectious organism target sequence. Generally, the probes will have at least 14 nucleotides, preferably at least 18 nucleotides, and more preferably at least 20 to 30 nucleotides of either of the complementary target nucleic acid strands and does not contain any hairpin secondary structures. In specific embodiments,

the probe can have a length of at least 30 nucleotides or at least 50 nucleotides. If there is to be complete complementarity, i.e., if the strand contains a sequence identical to that of the probe, the duplex will be relatively stable under even stringent conditions and the probes may be short, i.e., in the range of about 10-30 base pairs. If some degree of mismatch is expected in the probe, i.e., if it is suspected that the probe would hybridize to a variant region, or to a group of sequences such as all species within a specific genus, the probe may be of greater length (i.e., 15-40 bases) to balance the effect of the mismatch(es).

The probe need not span the entire SARS-CoV or the non-SARS-CoV infectious organism target gene. Any subset of the target region that has the potential to specifically identify SARS-CoV or the non-SARS-CoV infectious organism target or alelle can be used. Consequently, the nucleic acid probe may hybridize to as few as 8 nucleotides of the target region. Further, fragments of the probes may be used so long as they are sufficiently characteristic of the SARS-CoV or the non-SARS-CoV infectious organism target gene to be typed.

The probe or primer should be able to hybridize with a SARS-CoV or a non-SARS-CoV infectious organism target nucleotide sequence that is at least 8 nucleotides in length under low stringency. Preferably, the probe or primer hybridizes with a SARS-CoV or a non-SARS-CoV infectious organism target nucleotide sequence under middle or high stringency.

In still another aspect, the present invention is directed to an array of oligonucleotide probes immobilized on a support for typing a SARS-CoV or a non-SARS-CoV infectious organism target gene, which array comprises a support suitable for use in nucleic acid hybridization having immobilized thereon a plurality of oligonucleotide probes, at least one of said probes comprising a nucleotide sequence that: a) hybridizes, under high stringency, with a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17; or b) has at least 90% identity to a target SARS-CoV or a non-SARS-CoV infectious organism nucleotide sequence comprising a nucleotide

sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17.

The plurality of probes can comprise DNA, RNA, PNA or a derivative thereof. At least one or some of the probes can comprise a nucleotide sequence, or a complementary strand thereof, that is set forth in Table 13 or Tables 15-17. Preferably, probe arrays comprise all of the nucleotide sequences, or a complementary strand thereof, that are set forth in Table 13 or Tables 15-17. At least one, some or all of the probes can be labeled. Exemplary labels include a chemical, an enzymatic, an immunogenic, a radioactive, a fluorescent, a luminescent and a FRET label. Any suitable support, e.g., a silicon, a plastic, a glass, a ceramic, a rubber, and a polymer surface, can be used.

E. Assay formats

Immobilization of Probes

The present methods, probes and probe arrays can be used in solution. Preferably, it is conducted in chip format, *e.g.*, by using the probe(s) immobilized on a solid support.

The probes can be immobilized on any suitable surface, preferably, a solid support, such as silicon, plastic, glass, ceramic, rubber, or polymer surface. The probe may also be immobilized in a 3-dimensional porous gel substrate, *e.g.*, Packard HydroGel chip (Broude et al., *Nucleic Acids Res.*, 29(19):E92 (2001)).

For an array-based assay, the probes are preferably immobilized to a solid support such as a "biochip". The solid support may be biological, nonbiological, organic, inorganic, or a combination of any of these, existing as particles, strands, precipitates, gels, sheets, tubing, spheres, containers, capillaries, pads, slices, films, plates, slides, etc.

A microarray biochip containing a library of probes can be prepared by a number of well known approaches including, for example, light-directed methods, such as VLSIPS™ described in U.S. Patent Nos. 5,143,854, 5,384,261 or 5,561,071; bead based methods such as described in U.S. Patent No. 5,541,061; and pin based methods such as detailed in U.S. Patent No. 5,288,514. U.S. Patent No. 5,556,752, which details the

preparation of a library of different double stranded probes as a microarray using the VLSIPS™, is also suitable for preparing a library of hairpin probes in a microarray.

Flow channel methods, such as described in U.S. Patent Nos. 5,677,195 and 5,384,261, can be used to prepare a microarray biochip having a variety of different probes. In this case, certain activated regions of the substrate are mechanically separated from other regions when the probes are delivered through a flow channel to the support. A detailed description of the flow channel method can be found in U.S. Patent No. 5,556,752, including the use of protective coating wetting facilitators to enhance the directed channeling of liquids though designated flow paths.

Spotting methods also can be used to prepare a microarray biochip with a variety of probes immobilized thereon. In this case, reactants are delivered by directly depositing relatively small quantities in selected regions of the support. In some steps, of course, the entire support surface can be sprayed or otherwise coated with a particular solution. In particular formats, a dispenser moves from region to region, depositing only as much probe or other reagent as necessary at each stop. Typical dispensers include micropipettes, nanopippettes, ink-jet type cartridges and pins to deliver the probe containing solution or other fluid to the support and, optionally, a robotic system to control the position of these delivery devices with respect to the support. In other formats, the dispenser includes a series of tubes or multiple well trays, a manifold, and an array of delivery devices so that various reagents can be delivered to the reaction regions simultaneously. Spotting methods are well known in the art and include, for example, those described in U.S. Patent Nos. 5,288,514, 5,312,233 and 6,024,138. In some cases, a combination of flow channels and "spotting" on predefined regions of the support also can be used to prepare microarray biochips with immobilized probes.

A solid support for immobilizing probes is preferably flat, but may take on alternative surface configurations. For example, the solid support may contain raised or depressed regions on which probe synthesis takes place or where probes are attached. In some embodiments, the solid support can be chosen to provide appropriate light-absorbing characteristics. For example, the support may be a polymerized Langmuir Blodgett film, glass or functionalized glass, Si, Ge, GaAs, GaP, SiO_2, SiN_4,

modified silicon, or any one of a variety of gels or polymers such as (poly)tetrafluoroethylene, (poly)vinylidendifluoride, polystyrene, polycarbonate, or combinations thereof. Other suitable solid support materials will be readily apparent to those of skill in the art.

The surface of the solid support can contain reactive groups, which include carboxyl, amino, hydroxyl, thiol, or the like, suitable for conjugating to a reactive group associated with an oligonucleotide or a nucleic acid. Preferably, the surface is optically transparent and will have surface Si--OH functionalities, such as those found on silica surfaces.

The probes can be attached to the support by chemical or physical means such as through ionic, covalent or other forces well known in the art. Immobilization of nucleic acids and oligonucleotides can be achieved by any means well known in the art (*see, e.g.*, Dattagupta et al., *Analytical Biochemistry*, 177:85-89(1989); Saiki et al., *Proc. Natl. Acad. Sci. USA*, 86:6230-6234(1989); and Gravitt et al., *J. Clin. Micro.*, 36:3020-3027(1998)).

The probes can be attached to a support by means of a spacer molecule, *e.g.*, as described in U.S. Patent No. 5,556,752 to Lockhart et al., to provide space between the double stranded portion of the probe as may be helpful in hybridization assays. A spacer molecule typically comprises between 6-50 atoms in length and includes a surface attaching portion that attaches to the support. Attachment to the support can be accomplished by carbon-carbon bonds using, for example, supports having (poly)trifluorochloroethylene surfaces, or preferably, by siloxane bonds (using, for example, glass or silicon oxide as the solid support). Siloxane bonding can be formed by reacting the support with trichlorosilyl or trialkoxysilyl groups of the spacer. Aminoalkylsilanes and hydroxyalkylsilanes, bis(2-hydroxyethyl)-aminopropyltriethoxysilane, 2-hydroxyethylaminopropyltriethoxysilane, aminopropyltriethoxysilane or hydroxypropyltriethoxysilane are useful are surface attaching groups.

The spacer can also include an extended portion or longer chain portion that is attached to the surface-attaching portion of the probe. For example, amines, hydroxyl,

thiol, and carboxyl groups are suitable for attaching the extended portion of the spacer to the surface-attaching portion. The extended portion of the spacer can be any of a variety of molecules which are inert to any subsequent conditions for polymer synthesis. These longer chain portions will typically be aryl acetylene, ethylene glycol oligomers containing 2-14 monomer units, diamines, diacids, amino acids, peptides, or combinations thereof.

In some embodiments, the extended portion of the spacer is a polynucleotide or the entire spacer can be a polynucleotide. The extended portion of the spacer also can be constructed of polyethyleneglycols, polynucleotides, alkylene, polyalcohol, polyester, polyamine, polyphosphodiester and combinations thereof. Additionally, for use in synthesis of probes, the spacer can have a protecting group attached to a functional group (*e.g.*, hydroxyl, amino or carboxylic acid) on the distal or terminal end of the spacer (opposite the solid support). After deprotection and coupling, the distal end can be covalently bound to an oligomer or probe.

The present method can be used to analyze a single sample with a single probe at a time. Preferably, the method is conducted in high-throughput format. For example, a plurality of samples can be analyzed with a single probe simultaneously, or a single sample can be analyzed using a plurality of probes simultaneously. More preferably, a plurality of samples can be analyzed using a plurality of probes simultaneously.

Hybridization Conditions

Hybridization can be carried out under any suitable technique known in the art. It will be apparent to those skilled in the art that hybridization conditions can be altered to increase or decrease the degree of hybridization, the level of specificity of the hybridization, and the background level of non-specific binding (i.e., by altering hybridization or wash salt concentrations or temperatures). The hybridization between the probe and the target nucleotide sequence can be carried out under any suitable stringencies, including high, middle or low stringency. Typically, hybridizations will be performed under conditions of high stringency.

Hybridization between the probe and target nucleic acids can be homogenous, e.g., typical conditions used in molecular beacons (Tyagi S. et al., *Nature Biotechnology*, 14:303-308 (1996); and U.S. Patent No. 6,150,097) and in hybridization protection assay (Gen-Probe, Inc) (U. S. Patent No. 6,004,745), or heterogeneous (typical conditions used in different type of nitrocellulose based hybridization and those used in magnetic bead based hybridization).

The target polynucleotide sequence may be detected by hybridization with an oligonucleotide probe that forms a stable hybrid with that of the target sequence under high to low stringency hybridization and wash conditions. An advantage of detection by hybridization is that, depending on the probes used, additional specificity is possible. If it is expected that the probes will be completely complementary (i.e., about 99% or greater) to the target sequence, high stringency conditions will be used. If some mismatching is expected, for example, if variant strains are expected with the result that the probe will not be completely complementary, the stringency of hybridization may be lessened. However, conditions are selected to minimize or eliminate nonspecific hybridization.

Conditions those affect hybridization and those select against nonspecific hybridization are known in the art (Molecular Cloning A Laboratory Manual, second edition, J. Sambrook, E. Fritsch, T. Maniatis, Cold Spring Harbor Laboratory Press, 1989). Generally, lower salt concentration and higher temperature increase the stringency of hybridization. For example, in general, stringent hybridization conditions include incubation in solutions that contain approximately 0.1XSSC, 0.1% SDS, at about 65°C incubation/wash temperature. Middle stringent conditions are incubation in solutions that contain approximately 1-2XSSC, 0.1% SDS and about 50°C - 65°C incubation/wash temperature. The low stringency conditions are 2XSSC and about 30°C - 50°C.

An alternate method of hybridization and washing is first to carry out a low stringency hybridization (5XSSPE, 0.5% SDS) followed by a high stringency wash in the presence of 3M tetramethyl-ammonium chloride (TMAC). The effect of the TMAC is to equalize the relative binding of A-T and G-C base pairs so that the efficiency of

hybridization at a given temperature corresponds more closely to the length of the polynucleotide. Using TMAC, it is possible to vary the temperature of the wash to achieve the level of stringency desired (Wood et al., *Proc. Natl. Acad. Sci. USA*, 82:1585-1588 (1985)).

A hybridization solution may contain 25% formamide, 5XSSC, 5XDenhardt's solution, 100 μg/ml of single stranded DNA, 5% dextran sulfate, or other reagents known to be useful for probe hybridization.

Detection of the Hybrid

Detection of hybridization between the probe and the target SARS-CoV nucleic acids can be carried out by any method known in the art, *e.g.*, labeling the probe, the secondary probe, the target nucleic acids or some combination thereof, and are suitable for purposes of the present invention. Alternatively, the hybrid may be detected by mass spectroscopy in the absence of detectable label (e.g., U.S. Patent No. 6,300,076).

The detectable label is a moiety that can be detected either directly or indirectly after the hybridization. In other words, a detectable label has a measurable physical property (*e.g.*, fluorescence or absorbance) or is participant in an enzyme reaction. Using direct labeling, the target nucleotide sequence or the probe is labeled, and the formation of the hybrid is assessed by detecting the label in the hybrid. Using indirect labeling, a secondary probe is labeled, and the formation of the hybrid is assessed by the detection of a secondary hybrid formed between the secondary probe and the original hybrid.

Methods of labeling probes or nucleic acids are well known in the art. Suitable labels include fluorophores, chromophores, luminophores, radioactive isotopes, electron dense reagents, FRET(fluorescence resonance energy transfer), enzymes and ligands having specific binding partners. Particularly useful labels are enzymatically active groups such as enzymes (Wisdom, *Clin. Chem.*, 22:1243 (1976)); enzyme substrates (British Pat. No. 1,548,741); coenzymes (U.S. Patent Nos. 4,230,797 and 4,238,565) and enzyme inhibitors (U.S. Patent No. 4,134,792); fluorescers (Soini and Hemmila, *Clin. Chem.*, 25:353 (1979)); chromophores including phycobiliproteins, luminescers such as

chemiluminescers and bioluminescers (Gorus and Schram, *Clin. Chem.*, 25:512 (1979) and ibid, 1531); specifically bindable ligands, *i.e.*, protein binding ligands; antigens; and residues comprising radioisotopes such as ^{3}H, ^{35}S, ^{32}P, ^{125}I, and ^{14}C. Such labels are detected on the basis of their own physical properties (*e.g.*, fluorescers, chromophores and radioisotopes) or their reactive or binding properties (*e.g.*, antibodies, enzymes, substrates, coenzymes and inhibitors). Ligand labels are also useful for solid phase capture of the oligonucleotide probe (i.e., capture probes). Exemplary labels include biotin (detectable by binding to labeled avidin or streptavidin) and enzymes, such as horseradish peroxidase or alkaline phosphatase (detectable by addition of enzyme substrates to produce a colored reaction product).

For example, a radioisotope-labeled probe or target nucleic acid can be detected by autoradiography. Alternatively, the probe or the target nucleic acid labeled with a fluorescent moiety can detected by fluorimetry, as is known in the art. A hapten or ligand (*e.g.*, biotin) labeled nucleic acid can be detected by adding an antibody or an antibody pigment to the hapten or a protein that binds the labeled ligand (*e.g.*, avidin).

As a further alternative, the probe or nucleic acid may be labeled with a moiety that requires additional reagents to detect the hybridization. If the label is an enzyme, the labeled nucleic acid, *e.g.*, DNA, is ultimately placed in a suitable medium to determine the extent of catalysis. For example, a cofactor-labeled nucleic acid can be detected by adding the enzyme for which the label is a cofactor and a substrate for the enzyme. Thus, if the enzyme is a phosphatase, the medium can contain nitrophenyl phosphate and one can monitor the amount of nitrophenol generated by observing the color. If the enzyme is a beta-galactosidase, the medium can contain o-nitro-phenyl-D-galacto-pyranoside, which also liberates nitrophenol. Exemplary examples of the latter include, but are not limited to, beta-galactosidase, alkaline phosphatase, papain and peroxidase. For *in situ* hybridization studies, the final product of the substrate is preferably water insoluble. Other labels, *e.g.*, dyes, will be evident to one having ordinary skill in the art.

The label can be linked directly to the DNA binding ligand, *e.g.*, acridine dyes, phenanthridines, phenazines, furocoumarins, phenothiazines and quinolines, by direct

chemical linkage such as involving covalent bonds, or by indirect linkage such as by the incorporation of the label in a microcapsule or liposome, which in turn is linked to the binding ligand. Methods by which the label is linked to a DNA binding ligand such as an intercalator compound are well known in the art and any convenient method can be used. Representative intercalating agents include mono-or bis-azido aminoalkyl methidium or ethidium compounds, ethidium monoazide ethidium diazide, ethidium dimer azide (Mitchell et al., *J. Am. Chem. Soc.*, 104:4265 (1982))), 4-azido-7-chloroquinoline, 2-azidofluorene, 4'-aminomethyl-4,5'-dimethylangelicin, 4'-aminomethyl-trioxsalen (4'aminomethyl-4,5',8-trimethyl-psoralen), 3-carboxy-5- or -8-amino- or -hydroxy-psoralen. A specific nucleic acid binding azido compound has been described by Forster et al., *Nucleic Acid Res.*, 13:745 (1985). Other useful photoreactable intercalators are the furocoumarins which form (2+2) cycloadducts with pyrimidine residues. Alkylating agents also can be used as the DNA binding ligand, including, for example, bis-chloroethylamines and epoxides or aziridines, *e.g.*, aflatoxins, polycyclic hydrocarbon epoxides, mitomycin and norphillin A. Particularly useful photoreactive forms of intercalating agents are the azidointercalators. Their reactive nitrenes are readily generated at long wavelength ultraviolet or visible light and the nitrenes of arylazides prefer insertion reactions over their rearrangement products (White et al., *Meth. Enzymol.*, 46:644 (1977)).

The probe may also be modified for use in a specific format such as the addition of 10-100 T residues for reverse dot blot or the conjugation to bovine serum albumin or immobilization onto magnetic beads.

When detecting hybridization by an indirect detection method, a detectably labeled second probe(s) can be added after initial hybridization between the probe and the target or during hybridization of the probe and the target. Optionally, the hybridization conditions may be modified after addition of the secondary probe. After hybridization, unhybridized secondary probe can be separated from the initial probe, for example, by washing if the initial probe is immobilized on a solid support. In the case of a solid support, detection of label bound to locations on the support indicates hybridization of a target nucleotide sequence in the sample to the probe.

The detectably labeled secondary probe can be a specific probe. Alternatively, the detectably labeled probe can be a degenerate probe, *e.g.*, a mixture of sequences such as whole genomic DNA essentially as described in U.S. Patent No. 5,348,855. In the latter case, labeling can be accomplished with intercalating dyes if the secondary probe contains double stranded DNA. Preferred DNA-binding ligands are intercalator compounds such as those described above.

A secondary probe also can be a library of random nucleotide probe sequences. The length of a secondary probe should be decided in view of the length and composition of the primary probe or the target nucleotide sequence on the solid support that is to be detected by the secondary probe. Such a probe library is preferably provided with a 3' or 5' end labeled with photoactivatable reagent and the other end loaded with a detection reagent such as a fluorophore, enzyme, dye, luminophore, or other detectably known moiety.

The particular sequence used in making the labeled nucleic acid can be varied. Thus, for example, an amino-substituted psoralen can first be photochemically coupled with a nucleic acid, the product having pendant amino groups by which it can be coupled to the label, *i.e.*, labeling is carried out by photochemically reacting a DNA binding ligand with the nucleic acid in the test sample. Alternatively, the psoralen can first be coupled to a label such as an enzyme and then to the nucleic acid.

Advantageously, the DNA binding ligand is first combined with label chemically and thereafter combined with the nucleic acid probe. For example, since biotin carries a carboxyl group, it can be combined with a furocoumarin by way of amide or ester formation without interfering with the photochemical reactivity of the furocoumarin or the biological activity of the biotin. Aminomethylangelicin, psoralen and phenanthridium derivatives can similarly be linked to a label, as can phenanthridium halides and derivatives thereof such as aminopropyl methidium chloride (Hertzberg et al, *J. Amer. Chem. Soc.*, 104:313 (1982)). Alternatively, a bifunctional reagent such as dithiobis succinimidyl propionate or 1,4-butanediol diglycidyl ether can be used directly to couple the DNA binding ligand to the label where the reactants have alkyl amino residues, again in a known manner with regard to solvents, proportions and reaction

conditions. Certain bifunctional reagents, possibly glutaraldehyde may not be suitable because, while they couple, they may modify nucleic acid and thus interfere with the assay. Routine precautions can be taken to prevent such difficulties.

Also advantageously, the DNA binding ligand can be linked to the label by a spacer, which includes a chain of up to about 40 atoms, preferably about 2 to 20 atoms, including, but not limited to, carbon, oxygen, nitrogen and sulfur. Such spacer can be the polyfunctional radical of a member including, but not limited to, peptide, hydrocarbon, polyalcohol, polyether, polyamine, polyimine and carbohydrate, *e.g.*, -glycyl-glycyl-glycyl- or other oligopeptide, carbonyl dipeptides, and omega-amino-alkane-carbonyl radical or the like. Sugar, polyethylene oxide radicals, glyceryl, pentaerythritol, and like radicals also can serve as spacers. Spacers can be directly linked to the nucleic acid-binding ligand and/or the label, or the linkages may include a divalent radical of a coupler such as dithiobis succinimidyl propionate, 1,4-butanediol diglycidyl ether, a diisocyanate, carbodiimide, glyoxal, glutaraldehyde, or the like.

Secondary probe for indirect detection of hybridization can be also detected by energy transfer such as in the "beacon probe" method described by Tyagi and Kramer, *Nature Biotech.*, 14:303-309 (1996) or U.S. Patent Nos. 5,119,801 and 5,312,728 to Lizardi et al. Any FRET detection system known in the art can be used in the present method. For example, the AlphaScreen™ system can be used. AlphaScreen technology is an "Amplified Luminescent Proximity Homogeneous Assay" method. Upon illumination with laser light at 680 nm, a photosensitizer in the donor bead converts ambient oxygen to singlet-state oxygen. The excited singlet-state oxygen molecules diffuse approximately 250 nm (one bead diameter) before rapidly decaying. If the acceptor bead is in close proximity of the donor bead, by virtue of a biological interaction, the singlet-state oxygen molecules reacts with chemiluminescent groups in the acceptor beads, which immediately transfer energy to fluorescent acceptors in the same bead. These fluorescent acceptors shift the emission wavelength to 520-620 nm. The whole reaction has a 0.3 second half-life of decay, so measurement can take place in time-resolved mode. · Other exemplary FRET donor/acceptor pairs include Fluorescein

(donor) and tetramethylrhodamine (acceptor) with an effective distance of 55Å; IAEDANS (donor) and Fluorescein (acceptor) with an effective distance of 46Å; and Fluorescein (donor) and QSY-7 dye (acceptor) with an effective distance of 61Å (Molecular Probes).

Quantitative assays for nucleic acid detection also can be performed according to the present invention. The amount of secondary probe bound to a microarray spot can be measured and can be related to the amount of nucleic acid target which is in the sample. Dilutions of the sample can be used along with controls containing known amount of the target nucleic acid. The precise conditions for performing these steps will be apparent to one skilled in the art. In microarray analysis, the detectable label can be visualized or assessed by placing the probe array next to x-ray film or phosphoimagers to identify the sites where the probe has bound. Fluorescence can be detected by way of a charge-coupled device (CCD) or laser scanning.

Test samples

Any suitable samples, including samples of human, animal, or environmental (*e.g.,* soil or water) origin, can be analyzed using the present method. Test samples can include body fluids, such as urine, blood, semen, cerebrospinal fluid, pus, amniotic fluid, tears, or semisolid or fluid discharge, *e.g.*, sputum, saliva, lung aspirate, vaginal or urethral discharge, stool or solid tissue samples, such as a biopsy or chorionic villi specimens. Test samples also include samples collected with swabs from the skin, genitalia, or throat.

Test samples can be processed to isolate nucleic acid by a variety of means well known in the art (See generally, Ausubel (Ed.) *Current Protocols in Molecular Biology, 2. Preparation and Analysis of DNA and 4. Preparation and Analysis of RNA*, John Wiley & Sons, Inc. (2000)). It will be apparent to those skilled in the art that target nucleic acids can be RNA or DNA that may be in form of direct sample or purified nucleic acid or amplicons.

Purified nucleic acids can be extracted from the aforementioned samples and may be measured spectraphotometrically or by other instrument for the purity. For those

skilled in the art of nucleic acid amplification, amplicons are obtained as end products by various amplification methods such as PCR (Polymerase Chain Reaction, U.S. Patent Nos. 4,683,195, 4,683,202, 4,800,159 and 4,965,188), NASBA (Nucleic Acid Sequence Based Amplification, U.S. Patent No. 5,130,238), TMA (Transcription Mediated Amplification) (Kwoh et al., *Proc. Natl. Acad. Sci., USA*, 86:1173-1177 (1989)), SDA (Strand Displacement Amplification, described by Walker et al., U.S. Patent No. 5,270,184), tSDA (thermophilic Strand Displacement Amplification (U.S. Patent No. 5,648,211 and Euro. Patent No. EP 0 684315), SSSR (Self-Sustained Sequence Replication) (U. S. Patent No. 6,156,508).

In a specific embodiment, a sample of human origin is assayed. In yet another specific embodiment, a sputum, urine, blood, tissue section, food, soil or water sample is assayed.

Kits

The present probes can be packaged in a kit format, preferably with an instruction for using the probes to detect a target gene. The components of the kit are packaged together in a common container, typically including written instructions for performing selected specific embodiments of the methods disclosed herein. Components for detection methods, as described herein, may optionally be included in the kit, for example, a second probe, and/or reagents and means for carrying out label detection (*e.g.*, radiolabel, enzyme substrates, antibodies, etc., and the like).

F. Examples

Example 1. Probe designs

Various genome sequences of SARS-CoV are available (*See e.g.*, Table 22).

Table 22: Genome sequences of SARS coronaviruse currently obtained (as of 5/2/2003)

ID	Source of SARS	Submitting Country	GenBank Acc	Number of N in	Length of the	Percentage of N

	coronavirus e	(Area)		the sequence	genome	
SARS_BJ01	Beijing, China	China	AY278488	900	28920	3.11%
SARS_BJ02	Beijing, China	China	AY278487	300	29430	1.02%
SARS_BJ03	Beijing, China	China	AY278490	607	29291	2.07%
SARS_GZ01	Guangzhou, China	China	AY278489	1007	29429	3.42%
SARS_BJ04	Beijing, China	China	AY279354	2502	24774	10.10%
SARS_ CUHK-W1	Hong Kong, China	Hong Kong, China	AY278554	0	29736	0.00%
SARS_HKU-39849	Hong Kong, China	Hong Kong, China	AY278491	0	29742	0.00%
SARS_Urbani	Vietnam	U.S.	AY278741	0	29727	0.00%
SARS_TOR2	Toronto, Canada	Canada	AY274119	0	29736	0.00%

The sizes of the nine genomes shown in Table 22 are very similar. The five genomes submitted by China contain various levels of unidentified nucleotides (N).

The following Table 23 shows similarities or homologies among the nine genomes of SARS coronaviruse.

Table 23. Comparison of similarities between the nine genomes of SARS coronaviruse

	BJ01	BJ02	BJ03	GZ01	BJ04	CUHK-W1	HKU-39849	Urbani	TOR2
BJ01					91				
BJ02				94	88				
BJ03					89				
GZ01		94			91				
BJ04	91	88	89	91		89	89	89	89
CUHK-W1					89				
HKU-39849					89				
Urbani					89				
TOR2					89				

The similarity of the nine genomes of SARS coronaviruse were compared. The numbers shown in the Table 23 represent the percentage of similarity between two genomes. Each number in Table 23 equals to the number of the same bases in two genomes divided by the total number of bases (about 30,000 bases) compared and then timed by 100.

Table 23 shows that the different genomes of SARS coronaviruse are highly similar to each other except BJ04. The similarity lower than 99% is caused by the presence of N in the nucleotide sequence. If all the Ns in the nucleotide sequences from BJ01-BJ04 and GZ01 are considered as the same with other genome (this assumption is reasonable based on comparison of other part of the genomes), the nine genomes are 99% similar to each other.

Since SARS coronaviruse is conservative as shown in Tables 22 and 23, nucleic acid based detection methods are rational. Figure 1B indicates that detection of different parts of SARS coronaviruse genome at the same time can significantly increase the sensitivity and specificity of the detection method.

We have two overall designs. One design is to perform a multiplex PCR for different parts of SARS coronaviruse genome and use PCR products as probes for

detection. The second design is to perform a multiplex PCR for different parts of SARS coronaviruse genome and use a 70 mer oligonucleotides as probes for detection.

Target gene selection

Based on analysis of SARS coronaviruse genome, we selected three genes as target genes. These three genes are orf 1A and 1B polymerase proteins, spike protein, and nucleocapsid protein. We selected human housekeeping gene GAPD (glyceraldehyde 3-phosphate dehydrogenase) (GenBank Acc: NM_002046) as a positive control for RNA isolation. We selected a gene (*Arabidopsis*) (GenBank Acc: AJ441252), which has no homology to nucleotide sequence of human and common pathogens, as incorporated positive control.

Design of primers and probes

The three proteins of SARS coronaviruse were analyzed and their conservative sequences were compared. According to the requirement of multiplex PCR, multiple pairs of primers, which have similar Tm values and are 1.5 Kb in distance, and have amplified products between 200 to 900 bp, were designed based on the conservative sequence between different genomes. In addition, multiple non-overlapping oligonucleotides (70 mer) were designed based on amplified product of each pair of primers. These primers and probes were compared with the most updated NCBI nucleic acid non-redundant nucleotide database using BLASTN, and the specificities of the probes and primers were assured.

Example 2. Process for pretreatment of blood sample

Pretreatment of blood sample involves relatively complicated processes. However, considering the relative low concentration SARS virus in serum reported, pretreatment described herein can effectively enrich lymphocytes from about 2 ml of the whole blood in order to increase the chances of detection.

1. Sample collection and transfer

1) Samples collected from patients in the hospital room are put in a first transfer window. The door of the window is then closed and locked.

2) The samples are then transferred into a second transfer window. The samples are recorded in a notebook and three bar code labels are printed. The samples are tested for conventional detection and transferred into a pretreatment transfer window.

2. Use of biosafe cabinet

1) Hospital personnel for performing pretreatment process enters the pretreatment room and close the door. The biosafe cabinet is then turned on. The fan of the cabinet and light are then automatically turned on.

2) The indicator lights for power switch, air speed switch, and work light switch are checked for normal operation. The indicator light for air selection switch is checked as off status. Abnormal or unusual operation is reported.

3) The indicator light for alarm switch will make an alarm sound which indicates normal status of the biosafe cabinet after self-testing. Fifteen minutes later, the alarm sound from the indicator light for alarm switch is stopped and the process in the biosafe cabinet can be started.

4) The process in the cabinet cannot be started if the alarm sound continues or the process has to be stopped if there is an alarm sound during the process. The incident has to be reported immediately.

5) After the biosafe cabinet operates normally, samples are taken from the second transfer window and placed in the cabinet. The transfer window top is cleaned by wiping with 75% alcohol and spraying with 0.5% peracetic acid. The door for the transfer window is then closed and locked.

6) The complete process of sample pretreatment is then performed in the biosafe cabinet.

3. Serum isolation

1) Blood (1.8 ml) with anticoagulant is centrifuged for 10 minutes at 3,500 rpm. The top layer is marked with a marker pen.

2) The top layer serum (about 1.0 ml) is then collected and put into a 1.5 ml sterile Eppendorf centrifuge tube.

3) The Eppendorf centrifuge tube is labeled with the bar code (marked as "P") and labeled with a sequence number.

4) The sample is then recorded in a notebook.

5) The centrifuge tube containing the serum sample is put in a specialized sample box and stored at -80°C. The outside of the sample box is labeled with SARS, serum and range of sample numbers.

4. Isolation of blood cells

1) Lymphocyte isolation solution (3.6 ml) is added to a 10 ml centrifuge tube.

2) Sterile physiological saline (a volume equal to the serum taken out in the centrifuge tube described above) is added to the centrifuge tube containing the blood cells. The blood cells are resuspended in saline using Pasteur pipette.

3) The resuspended blood cells are slowly loaded on top of the lymphocyte isolation solution and centrifuged for 20 minutes at 1,500 rpm.

4) The cells located between the layers are collected and put in a 1.5 ml sterile Eppendorf centrifuge tube, which is then centrifuged for 5 minutes at 10,000 rpm to spin down the cells. The supernatant is withdrawn.

5) The tube containing the cell pellet is then labeled with the bar code (marked "C") and labeled with a sequence number.

6) The sample is recorded in a notebook.

7) The centrifuge tube containing the blood cell sample is put in a specialized sample box and stored at -80°C. The outside of the sample box is labeled with SARS, blood cells, and range of sample numbers.

8) The glass face plate of the biosafe cabinet is then opened. The bench surface and other surfaces in the biosafe cabinet are then sterilized by wiping with 70% alcohol and spraying 0.5% peracetic acid.

9) After cleaning, the glass face plate is closed. The ultraviolet light is placed inside the cabinet and turned on for 15 minutes.

10) The power switch of the biosafe cabinet is turned off before leaving the sample pretreatment room.

5. Matters needing attention

1) The lymphocyte isolation solution should not be used immediately after being taken out of the refrigerator. The solution should be used after its temperature reaches room temperature and the solution is mixed well.

2) The whole isolation process should be performed at 18-28°C. Too high or too low temperature can impact on the quality of isolation process.

3) The pipette tips, Eppendorf centrifuge tubes, gloves, and disposed reagents or liquids should be discarded in a waist tank (containing 0.5% peracetic acid). Everything in the waster tank should be treated at high pressure after experiment and then discarded.

4) 0.5% of peracetic acid is prepared by diluting 32 ml of 16% of peracetic acid in H_2O to make a final volume of 1,000 ml.

Example 3. Process for extracting RNA using QIAamp Viral RNA kit

The following procedures are used in RNA preparation:

1. Pipet 560 µl of prepared Buffer AVL containing Carrier RNA into a 1.5-ml microcentrifuge tube. If the sample volume is larger than 140 µl, increase the amount of Buffer AVL/Carrier RNA proportionally (e.g., a 280-µl sample will require 1120 µl Buffer AVL/Carrier RNA).

2. Add 140 µl plasma, serum, urine, cell-culture supernatant, or cell-free body fluid to the Buffer AVL/Carrier RNA in the microcentrifuge tube. Mix by pulse-vortexing for 15 sec. To ensure efficient lysis, it is essential that the sample is mixed thoroughly with Buffer AVL to yield a homogeneous solution. Frozen samples that have only been thawed once can also be used.

3. Incubate at room temperature (15–25°C) for 10 min. Viral particle lysis is complete after lysis for 10 min at room temperature. Longer incubation times have no effect on the yield or quality of the purified RNA. Potentially infectious agents and RNases are inactivated in Buffer AVL.

4. Briefly centrifuge the 1.5-ml microcentrifuge tube to remove drops from the inside of the lid.

5. Add 560 µl of ethanol (96–100%) to the sample, and mix by pulse-vortexing for 15 sec. After mixing, briefly centrifuge the 1.5-ml microcentrifuge tube to remove drops from inside the lid. Only ethanol is preferred since other alcohols may result in reduced RNA yield and purity. If the sample volume is greater than 140 µl, increase the amount of ethanol proportionally (e.g., a 280-µl sample will require 1120 µl of ethanol). In order to ensure efficient binding, it is essential that the sample is mixed thoroughly with the ethanol to yield a homogeneous solution.

6. Carefully apply 630 µl of the solution from step 5 to the QIAamp spin column (in a 2-ml collection tube) without wetting the rim. Close the cap, and centrifuge at 6000 x g (8000 rpm) for 1 min. Place the QIAamp spin column into a clean 2-ml collection tube, and discard the tube containing the filtrate. Close each spin column in order to avoid cross-contamination during centrifugation. Centrifugation is performed at 6,000 x g(8,000 rpm) in order to limit microcentrifuge noise. Centrifugation at full speed will not affect the yield or purity of the viral RNA. If the solution has not completely passed through the membrane, centrifuge again at a higher speed until all of the solution has passed through.

7. Carefully open the QIAamp spin column, and repeat step 6. If the sample volume is greater than 140 µl, repeat this step until all of the lysate has been loaded onto the spin column.

8. Carefully open the QIAamp spin column, and add 500 µl of Buffer AW1. Close the cap, and centrifuge at 6,000 x g (8,000 rpm) for 1 min. Place the QIAamp spin column in a clean 2-ml collection tube (provided), and discard the tube containing the filtrate. It is not necessary to increase the volume of Buffer AW1 even if the original sample volume was larger than 140 µl.

9. Carefully open the QIAamp spin column, and add 500 µl of Buffer AW2. Close the cap and centrifuge at full speed (20,000 x g; 14,000 rpm) for 3 min. Continue directly with step 10, or to eliminate any chance of possible Buffer AW2 carryover, perform step 9a, and then continue with step 10. Note: Residual Buffer AW2 in the eluate may cause problems in downstream applications. Some centrifuge rotors may vibrate upon deceleration, resulting in flow-through, containing Buffer AW2, contacting

the QIAamp spin column. Removing the QIAamp spin column and collection tube from the rotor may also cause flowthrough to come into contact with the QIAamp spin column. In these cases, the optional step 9a should be performed.

9a. (Optional): Place the QIAamp spin column in a new 2-ml collection tube (not provided), and discard the old collection tube with the filtrate. Centrifuge at full speed for 1 min.

10. Place the QIAamp spin column in a clean 1.5-ml microcentrifuge tube (not provided). Discard the old collection tube containing the filtrate. Carefully open the QIAamp spin column and add 60 µl of Buffer AVE equilibrated to room temperature. Close the cap, and incubate at room temperature for 1 min. Centrifuge at 6,000 x g(8,000 rpm) for 1 min. A single elution with 60 µl of Buffer AVE is sufficient to elute at least 90% of the viral RNA from the QIAamp spin column. Performing a double elution using 2 x 40 µl of Buffer AVE will increase yield by up to 10%. Elution with volumes of less than 30 µl will lead to reduced yields and will not increase the final concentration of RNA in the eluate. Viral RNA is stable for up to one year when stored at –20°C or –70°C.

The following are further information pertaining to the above procedures:

- Equilibrate samples to room temperature (15–25°C).
- Equilibrate Buffer AVE to room temperature for elution in step 10.
- Check whether Buffer AW1, Buffer AW2, and Carrier RNA have been prepared according to the instructions on pages 14–15.
- Redissolve precipitate in Buffer AVL/Carrier RNA by heating, if necessary, and cool to room temperature before use.
- All centrifugation steps are carried out at room temperature.

Example 4. An exemplary array format of SARS-CoV detection chip

Figure 5 illustrates an exemplary array format of SARS-CoV detection chip.

Immobilization control is an oligo-probe that is labeled by a fluorescent dye HEX on its end and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip.

Positive control(Arabidopsis) is an oligo-probe designed according to an *Arabidopsis* (one kind of model organism) gene and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip. During hybridization reaction, target probes that can hybridize with this positive control perfectly are added into the hybridization solution. Signals of the positive control can be applied to monitor the hybridization reaction.

Negative control is an oligo-probe that does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip.

Blank Control is DMSO solution spot. It is used for monitoring arraying quality.

SARS probes are 011, 024, 040 and 044 probes.

Example 5. SARS-CoV detection from a SARS patient blood sample (sample No. 3)

Figures 6A and 6B illustrate SARS-CoV detection from a SARS patient blood sample (sample No. 3). Lymphocytes were isolated from 3# SARS patient blood sample. RNA from lymphocytes was extracted by QIAamp Kit. RT-nest PCR was performed using RNA extracted above as templates. 044 RT-nest PCR result was good and hybridization result was good too. 040 RT-nest PCR result was poor but hybridization result was good. It shows that the chip-hybridization method is sensitive and specific.

Example 6. SARS-CoV detection from a SARS patient blood sample (sample No. 4)

Figures 7A and 7B illustrate SARS-CoV detection from a SARS patient blood sample (sample No. 4). Lymphocytes were isolated from 4# SARS patient blood sample. RNA from lymphocytes was extracted by QIAamp Kit. RT-nest PCR was performed using RNA extracted above as templates. 024, 040 and 044 RT-nest PCR results were good and hybridization results were good too.

Example 7. SARS-CoV detection from a SARS patient sputum sample (sample No. 5)

Figure 8 illustrates SARS-CoV detection from a SARS patient sputum sample (sample No. 5). RNA from 5# SARS patient sputum sample was extracted by QIAamp Kit. RT-nest PCR was performed using RNA extracted above as templates. 040 RT-nest PCR result was good and hybridization result was good too.

Example 8. SARS-CoV detection from a SARS patient sputum sample (sample No. 6)

Figure 9 illustrates SARS-CoV detection from a SARS patient sputum sample (sample No. 6). RNA from 6# SARS patient sputum sample was extracted by QIAamp Kit. RT-nest PCR was performed using RNA extracted above as templates. All probes RT-nest PCR results were good and hybridization results were good too.

Example 9. Another exemplary array format of SARS-CoV detection chip

Figure 10 illustrates another exemplary array format of SARS-CoV detection chip.

Immobilization control is an oligo-probe that is labeled by a fluorescent dye HEX on its end and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip.

Positive control (*Arabidopsis*) is an oligo-probe designed according to an *Arabidopsis* (one kind of model organism) gene and does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip. During hybridization reaction, target probes that can hybridize with this positive control perfectly are added into the hybridization solution. Signals of the positive control can be applied to monitor the hybridization reaction.

Negative control is an oligo-probe that does not participate in any hybridization reaction when a sample containing or suspected of containing of a SARS-CoV is contacted with the chip.

Blank Control is DMSO solution spot. It is used for monitoring arraying quality.

SARS probes are 011, 024, 040 and 044 probes.

Example 10. Possible positive results on the SARS-CoV detection chip illustrated in Figure 10

Figure 11 illustrates all possible positive results on the SARS SARS-CoV detection chip illustrated in Figure 10.

There are four sets probes on chips for detecting SARS virus: probe 011, probe 024, probe 040 and probe 044.

The first line gives the positive result (1) by signals appearing on all four sets of probes: 011+024+040+044.

The second line gives all the possible positive results (4) by signals appearing on three sets probes: 011+024+044, 024+040+044, 011+040+044, 011+024+040.

The third line gives all the possible positive results (6) by signals appearing on two sets probes: 011+040, 024+044, 011+044, 040+044, 011+024, 024+040.

The fourth line gives all the possible positive results (4) by signals appearing on only one set probes: 011, 024, 040, 044.

Example 11. Possible results on the SARS-CoV detection chip illustrated in Figure 12

Figure 13 illustrates all possible positive results on the SARS-CoV detection chip illustrated in Figure 12.

There are four sets of probes on chips for detecting SARS virus: probe 011, probe 024, probe 040 and probe 044.

The possible positive and negative results are also illustrated in Figure 14. The combinations for positive results include:

- 011 + 127;
- 040 + 127;
- 011 + 127 + 024;

- 011 + 127 + 044;
- 024 + 127 + 044;
- 011 + 127 + 024 + 040;
- 024 + 127;
- 044 + 127;
- 011 + 127 + 040;
- 024 + 127 + 040;
- 044 + 127 + 040;
- 011 + 127 + 044;
- 011 + 127 + 024 + 044;
- 011 + 127 + 024 + 040 + 044; and
- 127 + 024 + 040 + 044.

A negative result is indicated if only 127 is observed.

To be a valid assay result, positive or negative, the immobilization control signal (HEX) should always be observed.

The above examples are included for illustrative purposes only and are not intended to limit the scope of the invention. Many variations to those described above are possible. Since modifications and variations to the examples described above will be apparent to those of skill in this art, it is intended that this invention be limited only by the scope of the appended claims.